Studies on some soil-inhabiting arthropod fauna in the sugar beet crop.

Baker, A. N.

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STUDIES ON SOME SOIL-INHABITING ARTHROPOD FAUNA

IN THE SUGAR-BEET CROP

Submitted by A.N. BAKER B.Sc.

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FRONTISPIECE

SOME SOIL-INHABITING PESTS OF SUGAR-BEET

SEEDLINGS : a - BLANIULUS GUTTULATUS (BOSC.)

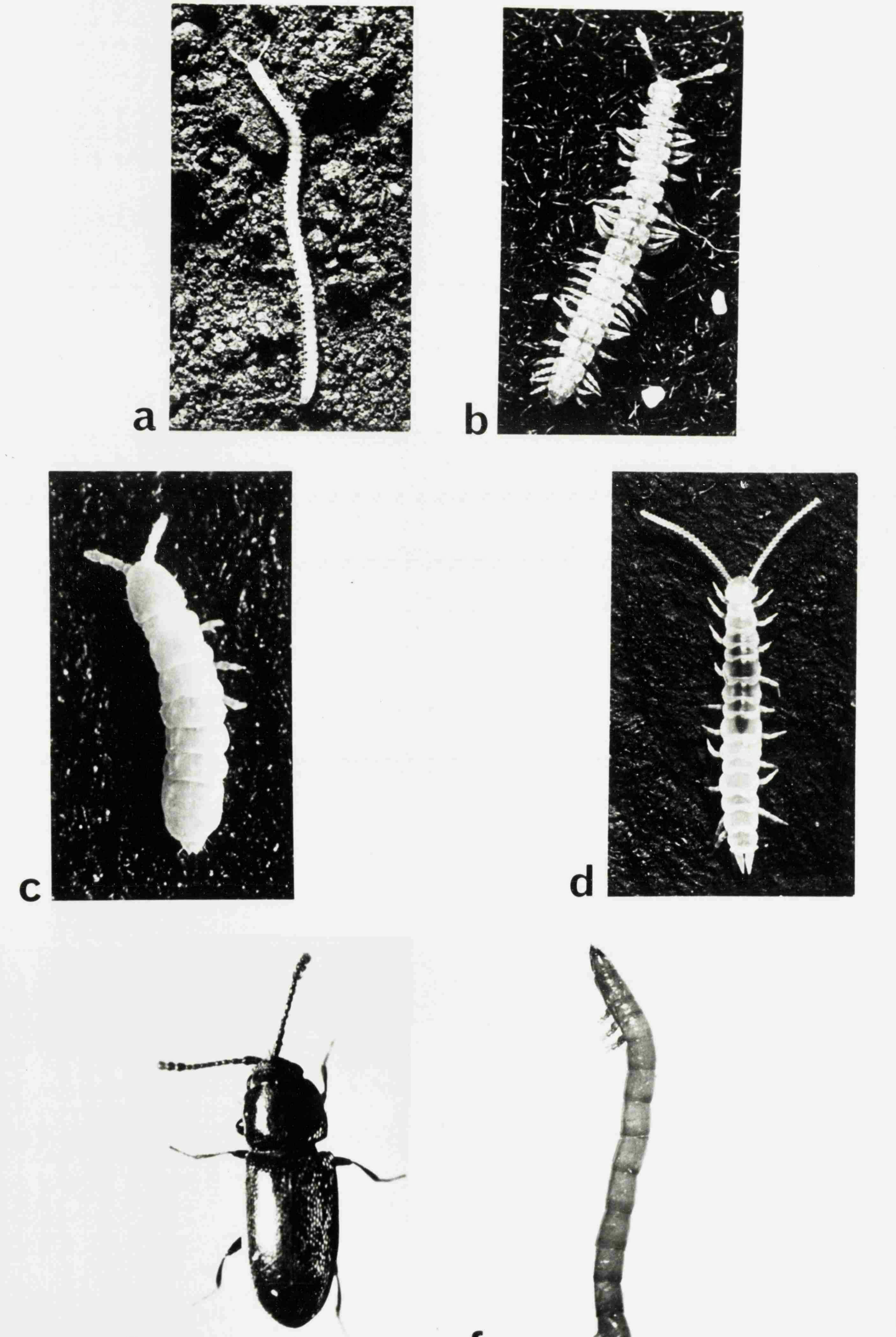
b - BRACHYDESMUS SUPERUS LATZEL

c - ONYCHIURUS ARMATUS (TULLBERG)

d - SCUTIGERELLA IMMACULATA NEWPORT

e - ATOMARIA LINEARIS STEPHENS

f - AGRIOTES SP. ESCHSCHOLTZ


a
b
c
d
e
f

SUMMARY

A four year study investigated some soil-inhabiting pests which cause damage to sugar-beet seedlings. Sampling methods are discussed and a convenient and quick flotation technique for extracting arthropods from soil samples, and other methods, are described.

Soil sampling sugar-beet fields in Spring, at many pest-infested sites, showed that blaniulid millipedes, pygmy beetle (*Atomaria linearis*) and onychiurid Collembola migrate from the subsoil and inter-row spaces and move progressively into the rows where they aggregate around sugar-beet seedling roots.

'Drilling-to-a-stand', with the concomitant wider seed spacing, sometimes increased numbers of millipedes and pygmy beetles per seedling root-zone, but never those of onychiurid Collembola. ?
Seedling roots at 9 in. spacing were usually damaged more than those from seed spaced at either 4.5 in. or 1.5 in. ~~but~~ and the narrowest spacing sometimes improved seedling growth. Seedlings were smaller on plots receiving a pre-emergence herbicide but numbers of ? root-zone pests and damage ~~was~~ were not affected. The damage to seedlings caused by millipedes is described in detail; it was also confirmed that *Onychiurus* spp. Collembola could kill seedlings in the laboratory and were generally the most common soil-inhabiting pest.

Some factors which may affect damage by soil-inhabiting pests in the field and laboratory were reported. Timing of sowing could affect seedling survival; raw and pelleted seed

was differentially attractive to some pests; soil compaction was sometimes beneficial.

The biology and life-histor~~y~~ies of millipedes (*Brachydesmus superus*) and two common blaniulids: *Blaniulus guttulatus* and *Boreoiulus tenuis* ~~was~~ were studied both in the field and in the laboratory. Two autumn surveys plotted the distribution of millipedes in the major sugar-beet growing areas in England. not millipedes

Some aspects of millipede behaviour ~~was~~ were studied in the laboratory; soil temperature ~~levels~~ affected both their orientation and pattern of feeding behaviour; vertical and horizontal soil columns tested their reaction to soil moisture status. effect?

Studies on the effect of different insecticides showed that seed-treatments may be effective in the laboratory but their performance in the field, against millipedes, was inconsistent. Many insecticides, in seed or furrow formulations, killed pygmy beetle; the currently recommended insecticide, gamma-BHC, performed consistently well but was toxic to a mite predator of *Onychiurus*.

1. GENERAL INTRODUCTION

In the United Kingdom, about 35% of the total sugar consumed is from the home-grown sugar-beet crop of 500,000 acres; ~~it~~ sugar beet thus occupies a prominent place in the farming economy. Sugar beet has been grown in England on a commercial scale since 1911 and there are now 17 sugar factories incorporated into the British Sugar Corporation (B.S.C.), most of them situated in eastern England where there is a concentration of arable farming; beet is grown in rotation with other crops, usually cereals and potatoes. The crop is profitable and growers know in advance the price to be paid for it; in 1971 a total of £61 million was paid for the beet delivered to the factories (Rose, 1972). As it is such a high value crop growers are given special assistance in obtaining the maximum sugar yield. Fieldmen, employed by the B.S.C., advise on all aspects of growing and ensure that conditions of the contract between factory and grower are fulfilled for both to receive the maximum benefit.

One of the reasons for poor yields is loss are losses due to pest attack; pests cause most damage in April and May (Dunning, 1975), when the sugar beet are still in the seedling stage (sugar beet are considered to be 'seedlings' up to about the four rough-leaf stage). The sugar-beet seedling is particularly susceptible to injury because it only has one axial growing point in its shoot and root system and is unable to 'tiller', like cereals, to produce new shoots or roots. The young, soft tissue of the single root is easily damaged by root-feeding pests (Jones and Dunning, 1972).

Economic damage by seedling pests is not as great as that by aphids spreading viruses. For instance, in 1974, Virus Yellows damage exceeded £10 million but this was exceptional and followed ten years of comparatively mild attacks. Seedling pests are less dramatic but more consistent in causing annual damage. As reported by Dunning (1975), in the four years 1965-69, millipedes, slugs, symphylids, wireworms and pygmy beetle caused annually, losses averaging £53,000. This figure was calculated on estimated loss of £100/acre for crop failure, £50 for severe damage, £25 for moderate damage and £10 for slight damage. The figure is probably an underestimate since much damage underground remains unnoticed and hence not reported. By In contrast, the readily observable foliage damage and particularly the yellowing of the foliage, symptomatic of virus attack, lends itself to a more accurate assessment. A few of the reports of increasing yearly damage by lesser known soil-inhabiting pests are probably just as much a consequence of the increased awareness of the observer as to any true increase in incidence. However, the role of the soil biologist is to search for unsuspected pests and understand their biology and behaviour. no caps.

Sugar-beet research aims at improving all aspects of its economy. Entomological research on seedling pests is usually directed towards the protection of the seedling crop and maintenance of a regular, vigorous, seedling population. The main root-feeding pests belong to ~~derive~~ from those arthropod groups which are mainly either entirely soil-inhabiting or soil surface-inhabiting (epigeic soil fauna). Most studies of soil fauna have concentrated on bare soil, grass or woodland litter. Insufficient attention has been given to the study of the economically important soil inhabitants within a specific crop. These are both the creatures which are phytophagous and have biting mouth parts which can become pests and those which are recognised as being beneficial because they prey on pests of economic importance. can they

One of the main objectives was ~~the~~ to study ~~of~~ the soil-inhabiting pests of sugar-beet seedlings. Other studies covered the wider spectrum of soil biology concerned with the general arthropod soil fauna of sugar-beet fields with particular reference to those which are most commonly found in the early stages of sugar-beet growth.

2. BACKGROUND TO STUDIES ON SOIL-INHABITING PESTS

The modern trend in sugar-beet growing is towards the replacement of labour-intensive stages in the growing of the crop with the use of modern agricultural techniques (Plate 1). Sugar beet is being sown at increasingly wider seed spacings using precision drilling methods and pelleted, genetic monogerm seed. The decreasing use of thinning operations and increasing use of pre-emergence herbicide has drastically reduced the need for hand labour in the early stages. There is now more concern about seedling damage since there are fewer seedlings present and hence, small losses are more noticeable.

Petherbridge and Stapley (1935) were the first to survey and record the incidence of pest damage in England and Wales. Regular monthly reporting of the incidence of damage by pests was investigated in 1947 and from 1957 until the present time a direct comparison of the importance of a wide range of pests is available from B.S.C. survey reports (Dunning, 1975). The data on severity of pest attack is in the form of the number of acres damaged in four categories: acres failed, severely, moderately and slightly affected; this is converted to an Index based on a Logarithmic scale which can be used for comparative purposes. Plate 2 illustrates the changing pattern of importance of two common soil-inhabiting seedling pests, wireworms and millipedes, with their respective Indices for the period 1958-1974. Whereas wireworm damage is shown to be generally decreasing, perhaps reflecting the efficiency of control measures, that due to millipedes appears to be increasing in importance though there is considerable variation from year to year. The incidence of

PLATE 1. PROGRESS IN THE ESTABLISHMENT OF SUGAR-BEET CROPS IN GREAT BRITAIN (1958 - 73)

Progress in the establishment of sugar beet crops in Great Britain (1958-73)

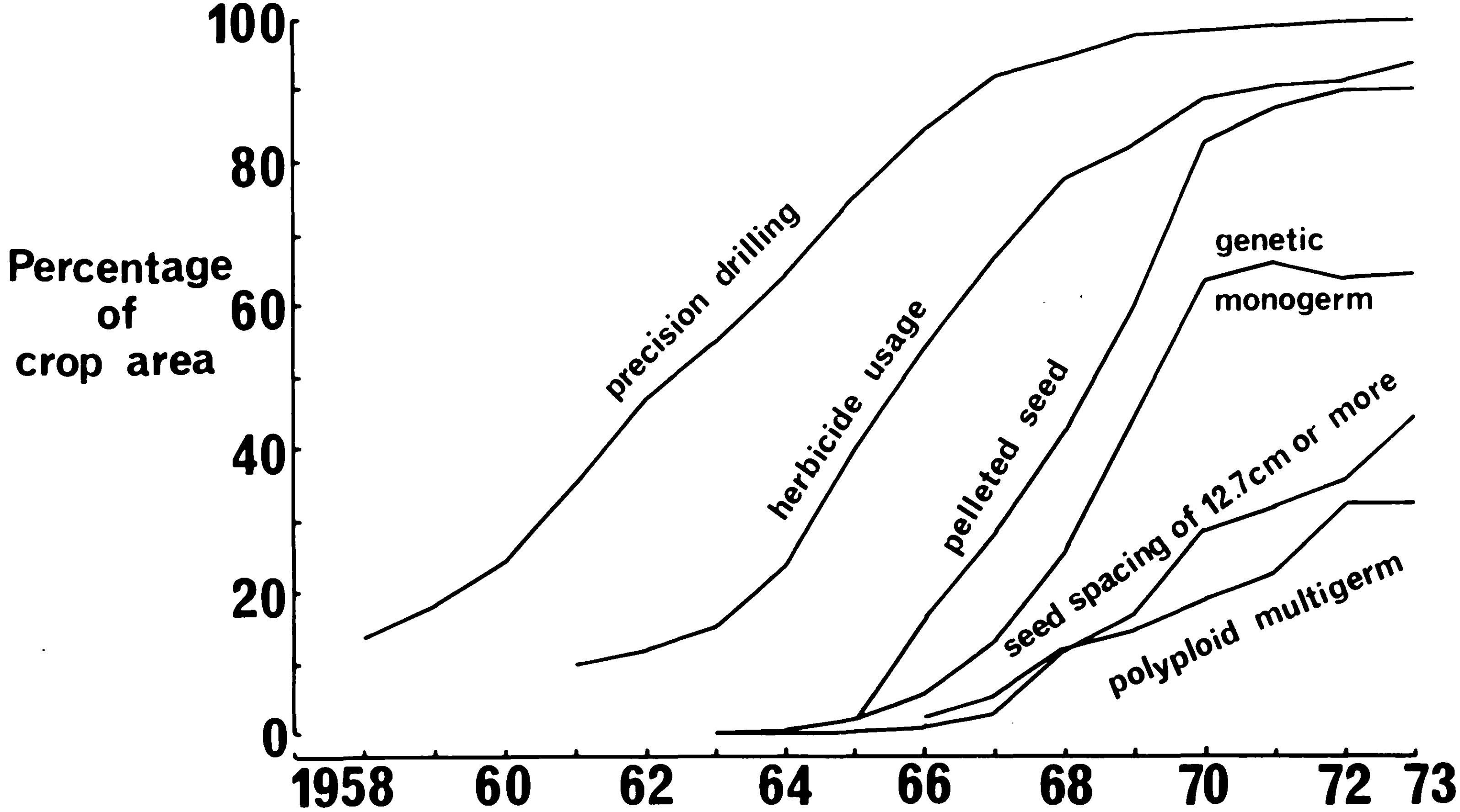
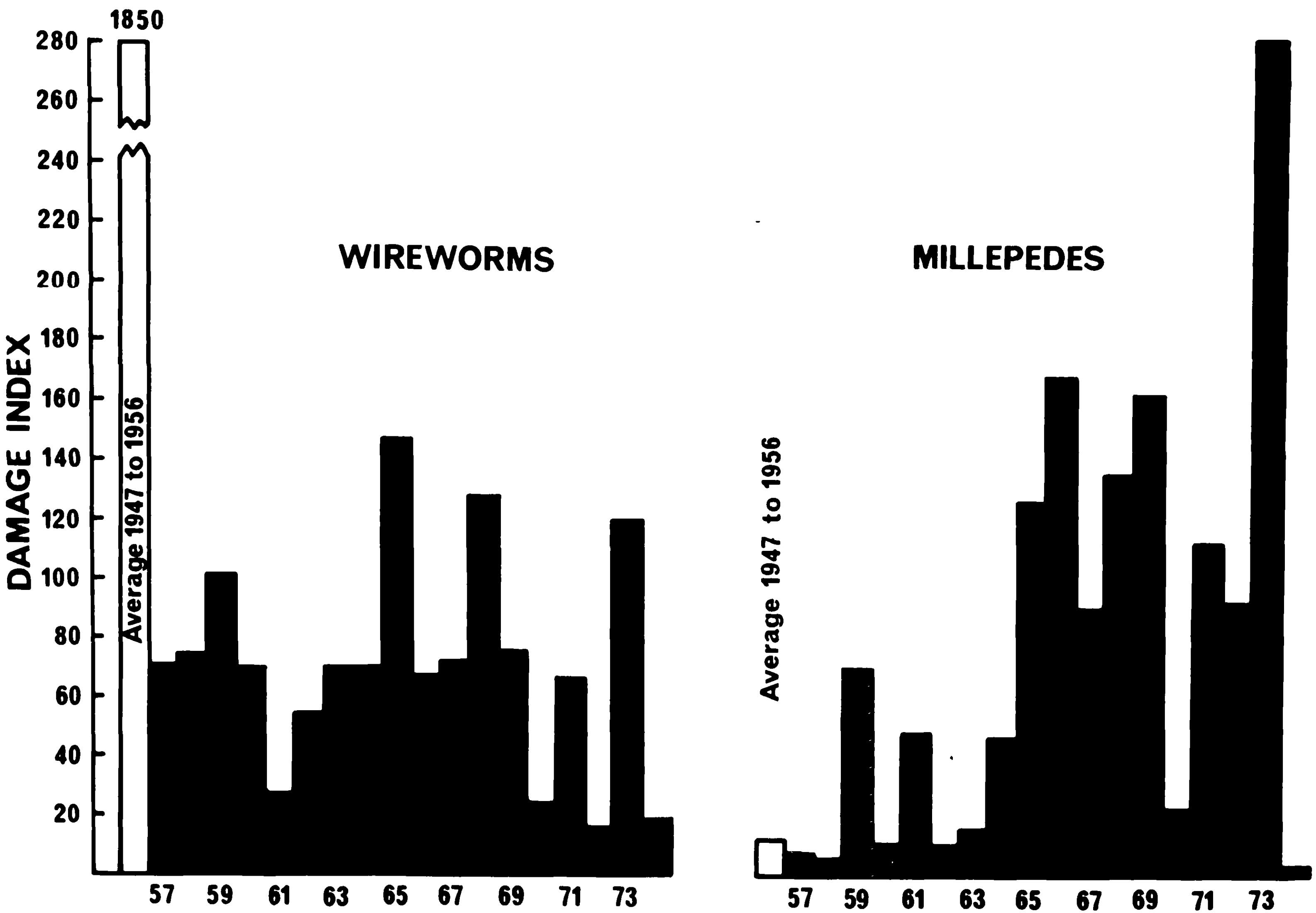


PLATE 2. <u>THE COMPARATIVE IMPORTANCE OF WIREWORMS AND MILLIPEDES FROM 1957 - 1974</u>.


1850
DAMAGE INDEX
280
260
240
220
200
180
160
140
120
100
80
60
40
20
Average 1947 to 1956
WIREWORMS
57
59
61
63
65
67
69
71
73
MILLEPEDES
Average 1947 to 1956
57
59
61
63
65
67
69
71
73

damage by other soil-inhabiting pests such as Symphylids, first recognised in 1965, is also increasing. It is suspected that the general increase in reported incidence of seedling damage by soil-inhabiting pests may be due to the increasing adoption of precision drilling methods and possibly linked with the now almost 100% use of herbicides in the crop. In France and Belgium, millipedes ~~have~~ are ~~assumed the position of~~ the most important pest of seedling beet whereas in Holland soil-inhabiting Collembola (Onychiurus sp.) are a particularly serious pest and may be causing unsuspected damage in England. The typical pattern of damage caused by such soil-inhabiting pests is illustrated in Plate 3 which shows a particularly severe attack by millipedes in a field with sugar beet. The seedling population, through the feeding activities of the pest, has been affected to the extent that a re-sowing would be necessary since the remaining seedlings would not be able to grow sufficiently large for their yield to compensate for their low numbers. Growers aim at a plant population of 30,000 per acre which, when regularly spaced, gives the best sugar yield. Emphasis must be placed on 'regular' since pest attacks not only cause growth retardation but create irregular gaps and bare patches within the field which probably reflect the distribution of the pest. Biernaux (1966) recorded a 22% reduction in root weight and a 25% loss in the weight of sugar from a crop that had been attacked by millipedes. Most seedlings that are attacked have their growth checked temporarily and effects are difficult to estimate but usually poor yields are the result of an irretrievable loss of seedlings. Although losses due to soil-inhabiting pests are relatively small on a national scale, they can represent a considerable problem for growers in some areas. Since pests such as millipedes have relatively little mobility, infestations

why probably?

PLATE 3. THE EFFECT OF AN INFESTATION BY MILLIPEDES, *BLANIULUS GUTTULATUS*, ON THE POPULATION OF SUGAR-BEET SEEDLINGS IN A FIELD AT MAGDALEN (NORFOLK) IN 1971.

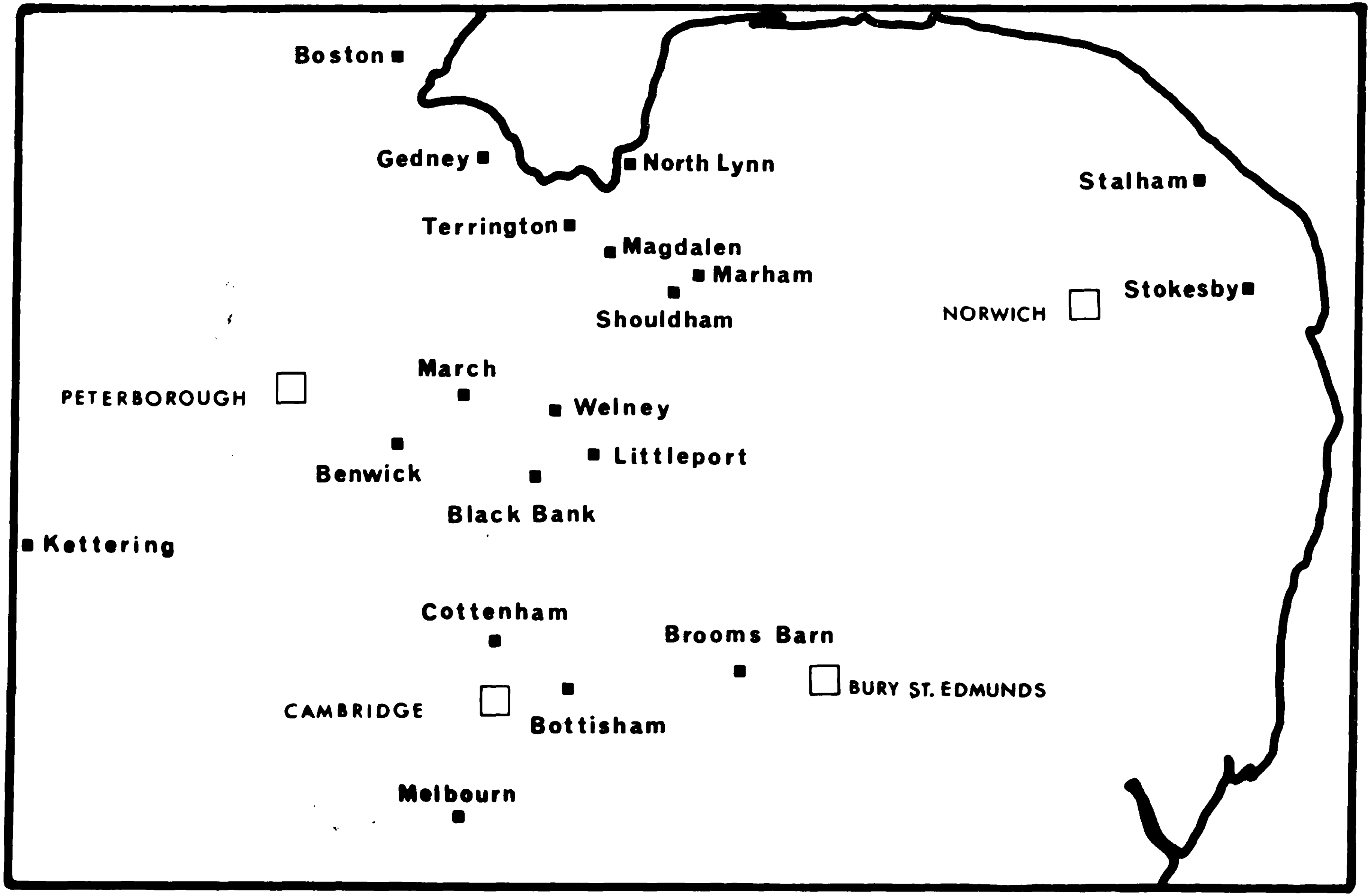
tend to be localised and it is noted that they can reappear in the same area of a field even after a four year rotation break. The incidence of damage by many pests, especially millipedes and pygmy beetle, is very high in some areas, especially in the fenland and areas bordering the western edge of the Wash (Dunning, 1975). Here, growers incur considerable annual expense in treating the soil with insecticides in addition to using the standard dieldrin-treated seed.

The increasing importance of the soil-inhabiting group of pests has highlighted the need for more intensive research into the effects of modern husbandry methods and has given the incentive to develop methods of increasing seedling protection, whether by chemical or cultural means. Although most studies involved a wide spectrum of pests the more detailed ones concentrated on millipedes, since they are the most difficult to control by existing methods and are a group whose economic importance has been neglected relative to most other pests. It was felt that investigations into some aspects of both their biology and behaviour might ellucidate their movements in the field and explain their apparent 'elusiveness'.

You or others

Studies involved mostly field experiments on commercial farms throughout East Anglia; Plate 4 shows the geographical location of experiments and where observations were made on pest-infested sites which are referred to in later chapters. Experiments were conducted with the co-operation of the staff of the British Sugar Corporation on fields where sugar beet was grown in normal rotation, though sometimes special permission was needed for the growing of two consecutive sugar-beet crops. Normal cultivation procedures were adhered to but often seed was sown either without insecticide or

PLATE 4. GEOGRAPHICAL LOCATION OF PEST-INFESTED SITES 1970 - 1974.



Boston
Gedney
North Lynn
Stalham
Terrington
Magdalen
Marham
Shouldham
NORWICH
Stokesby
March
PETERBOROUGH
Welney
Benwick
Littleport
Black Bank
Kettering
Cottenham
Brooms Barn
BURY ST. EDMUNDS
CAMBRIDGE
Bottisham
Melbourn

with one of a range of different compounds. Observations and measurements in the crop were carried out usually from the time of emergence until the seedlings were sufficiently large to be singled.

Results of experiments were usually sent for analysis to the Statistics Department at Rothamsted Experimental Station where they were transferred to punched tape and analysed by Analysis of Variance on the computer.

3. METHODS USED FOR SAMPLING AND EXTRACTING SOIL-INHABITING ARTHROPODS

3.1. INTRODUCTION

In the present study the range of soil animals to be sampled and recorded varied from small Collembola and mites sometimes less than 0.1 in. to millipedes up to 1.0 in. long; also included were soft bodied creatures such as Symphyla, and hard bodied Coleoptera. Graham and Stark (1954) mentioned that 'the sampling of a particular insect population must be resolved about the distribution and life-cycle of the insect involved'. Clearly this is difficult to apply in a study of such a wide spectrum of soil animals, especially as it involves different types of habitat; one of the main topics of study was to establish how soil pests react to the presence of sugar-beet seedlings which involved soil sampling at intervals both in bare soil and assessing their numbers around the roots of growing seedlings.

The problem of the most appropriate method of extracting the creatures from the soil samples was also not easily resolved owing to the behavioural and physical differences of the most important groups. The presence of non-active stages and the considerable variation in mobility of different animals, particularly of millipedes, largely precluded the general use of a dry extractor (where the animals are made to leave the substrate under some applied stimuli). However, where a chemical treatment was sampled and it was necessary to record both live and dead individuals, a 'dry extraction' apparatus was sometimes used or a flotation method which did not kill the animals.

In this section the stages involved in the extraction of soil animals: collection of the sample from the field; separation of animals from soil and assessment and identification used in the present study, are described and discussed.

3.2. COLLECTION OF THE SAMPLE

Plate 5 shows the different augers used to collect samples of soil-inhabiting creatures and Table 1 gives the specification of each.

The first, 'A', used for all soil sampling in 1970 only, was not used extensively; in moist soil it proved unsatisfactory because of the tendency of the soil to adhere to the inside wall making core removal difficult. In 1971 it was decided that the maximum sampling depth would be 3.9 in (10 cm.) since up to this rooting size seedlings are most susceptible to damage, most seedlings sampled had smaller roots. The stage at which they were sampled varied from the time of the appearance of the cotyledons above the soil surface until the two-leaf stage. Auger 'B' was used in 1971 and 1972 for taking samples centred on and including seedlings from seed spacing experiments, where seedlings were either 3, 4.5 or 9 in. apart and on chemically treated rows where a 3 in. spacing was used. It was also used until 1973 for sampling

PLATE 5. <u>AUGERS USED FOR SAMPLING SOIL-INHABITING ARTHROPODS</u>. See text for details.

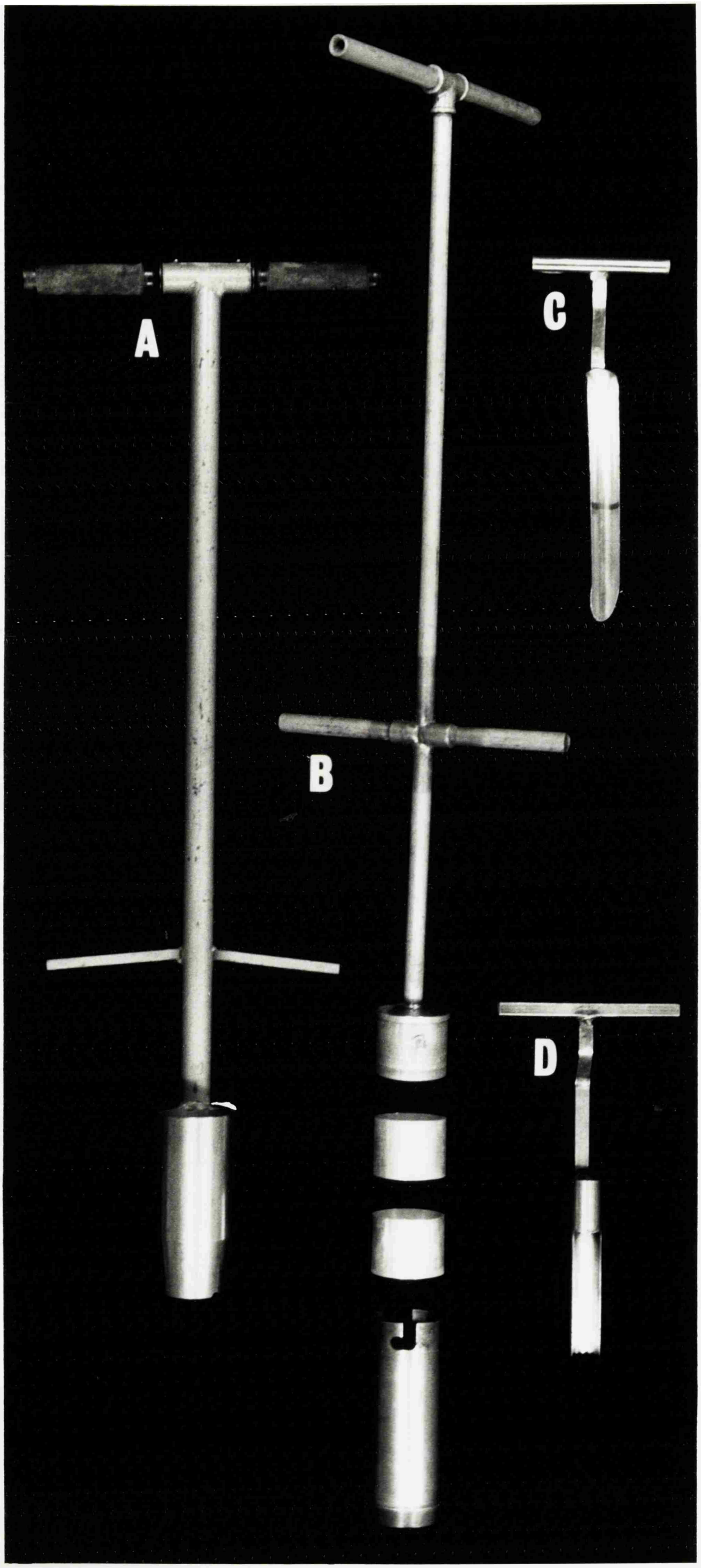

A
B
C
D

TABLE 1 SPECIFICATION OF AUGERS USED IN SOIL SAMPLING

Type	Auger diameters (in.)		Depth (in.)	Usable depth (in.)	Operation
	cutting	internal			
A	2.0	2.3	6.0	11.0	hand/foot
B	2.3	2.5	3.9	16.0	hand/foot
C	1.0	1.0	3.9	9.0	hand
D	1.0	1.0	3.9	8.0	hand

bare soil. This auger featured removable aluminium liners, similar to that used by Macfadyen (1961). The soil core, 2 x 1.9 in. deep, was easily removed from the auger by first removing the handle and plug, inverting the tool, cutting a 0.5 in. core of soil away from within the cutting edge and pressing the remaining 3.9 in. core out into a plastic bag. It was successful in relatively moist soil; if the soil was insufficiently moist to be retained, the liners were omitted. The auger was originally designed so that each of the 1.9 in. deep liners would fit into a dry, high-gradient canister-type apparatus (Macfadyen, 1961). The long handle made it suitable for sampling arthropods at different depths; each sample was a vertical succession of cores 3.9 in. deep using a 3.9 in. liner.

~~On~~ In seed spacing experiments the large sample variance necessitated ~~an increase in the number of~~ more units per treatment. Since most sampling involved taking cores centred on individual seedlings then the diameter of the core needed only to be sufficiently large to include the pests feeding on or aggregating in close contact with the root system. Observations in the field suggested that the volume enclosed within a 0.8 in. radius of the seedling would be sufficient and would be a more suitably sized core for enclosing seedlings at 1.5 in. spacing than for previous augers. Moreover, the number of sample units could be increased conveniently without a corresponding increase in sample weight, thus improving the accuracy of the sample by decreasing the sample variance. An auger recommended for sampling eelworm populations was tested in 1972. It consisted of a stainless-steel half-cylindrical sampler, diameter 1.5 in., depth 8 in. (auger 'C') with a mark at the 3.9 in. depth. Sample core units thus taken on each plot were bulked together to form 'bulked units' each comprising ten cores.

In 1973 and 1974 a three-quarter round auger, 1 in. diameter, cutting a core 3.9 in. deep, was used. It featured teeth on the cutting edge to facilitate penetration in hard, compacted soil, and it cut a more uniform volume of soil since less tended to fall away on extracting the auger from the hole. Again, sample cores from a single plot were combined to form bulked units of five or eight cores. A limitation on the number of cores was imposed by the total number of seedlings which could be sampled, and if the plot was sampled on successive dates a compromise had to be reached between accuracy and the number of seedlings available. The latter was determined by plot size which was usually as large as the space available permitted.

In bare soil there was no constraint on the number of cores that could be taken other than that dictated by cost and time needed for extraction. The auger used most frequently ('B') cut cores of 2.3 in. diameter. ~~For different workers interested in the population of a particular group within the soil fauna~~, sample size varied depending on the size of the animal to be extracted. Debauche (1962) stated that, whereas for Acarina and most Collembola, sampling units with a volume of 50-100 cm^3 were convenient, it was too small for animals from groups such as the Lumbricidae, Isopoda, Diplopoda, and Coleoptera. Morris (1927), estimating the total fauna in arable plots, took between 5 and 7 samples per treatment, each composed of a 9 in. cube of soil. Many soil-inhabiting pests such as Symphyla (Salt et al, 1948) and Onychiurus spp. (Glasgow, 1939) are non-randomly distributed and therefore an increase in the number of sample cores will increase the accuracy of a population estimate. Edwards and Dennis (1962) took 20 samples 2.5 in. diameter in a vertical succession of 2.5 in. for estimating Symphyla numbers in the soil. In the present studies the same criteria can be applied and usually between 16 and 48 cores were taken at any one depth to represent the original seed-bed estimate. Details of the number and size of samples used to estimate pest numbers are given later in Chapter 4.

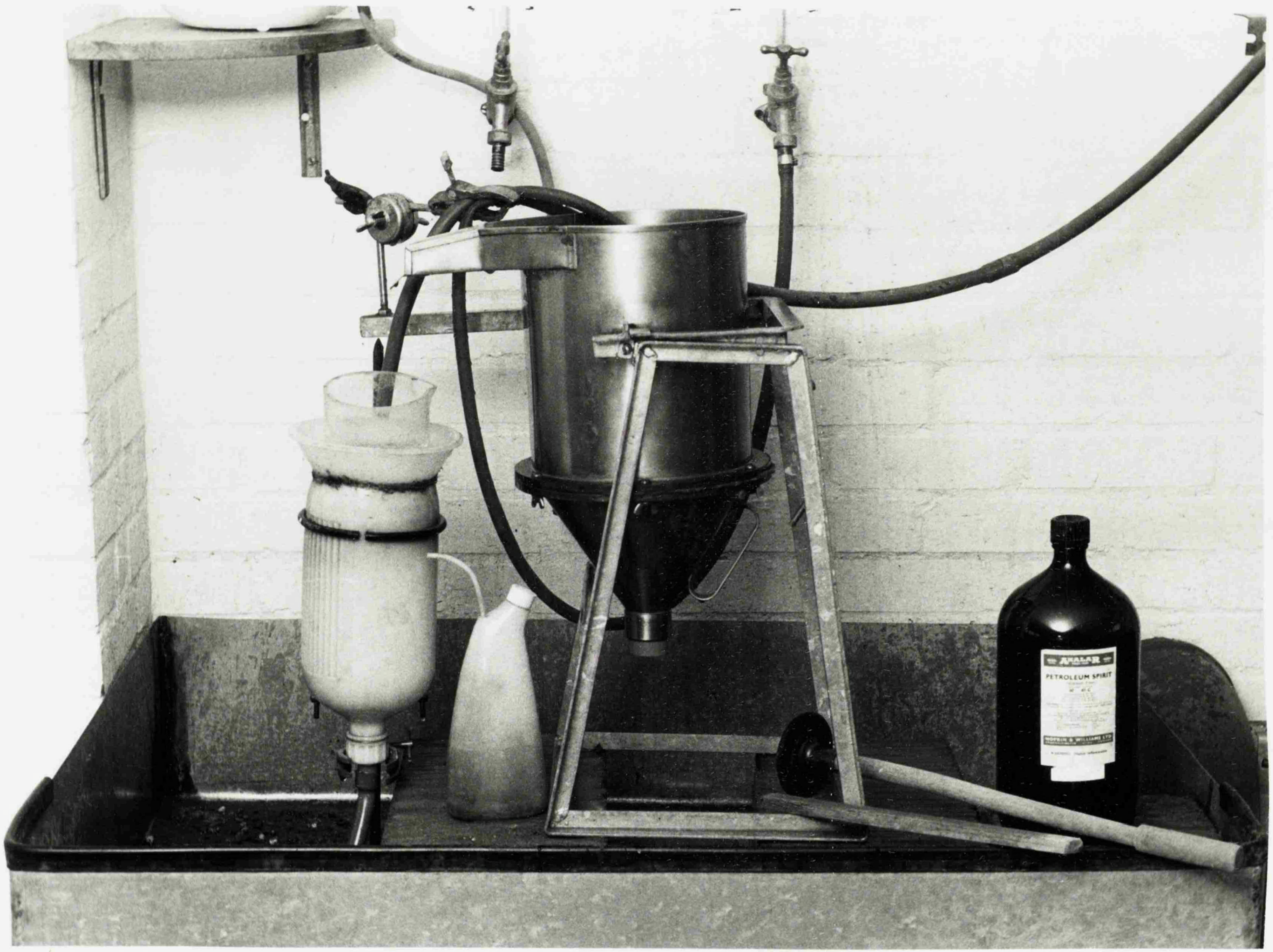
3.3. SEPARATION OF ARTHROPODS FROM SOIL SAMPLES

The choice of extraction method depended on both the arthropods that were of most importance and the substrate to be sampled. Since most problems in the establishment of seedlings concerned infestation by millipedes, this group was considered to be of prime interest, especially since relatively few workers have extracted them from arable soil. Hand-sorting methods used by Biernaux and Baurant (1964b) were too laborious for the processing of many samples from experiments; dry extraction methods, whilst suitable for litter analysis of woodland floors (Blower and Gabbut, 1964), have not been used fortotal estimates of millipedes in arable land. Murphy (1962) conjectured that the comparatively large size of Diplopoda may prohibit their efficient extraction by conventional funnel-type extractors and the alternative, the controlled draught funnel designed by Macfadyen (1953) which was used successfully to recover Glomeris sp., was too large for the present studies. Millipedes were recovered in large numbers by a wet sieving method used by Morris (1922, 1927) in his census of invertebrate fauna of arable land whilst Salt et al (1948) used a high density liquid, magnesium sulphate, to separate millipedes from soil. Since millipedes are sometimes either in a temporarily inactive condition (e.g. when moulting), or may be present as relatively immobile stadia, a flotation method is likely to be the most accurate method to extract all stages in a population study. Macfadyen (1953) found that a flotation method (modified Salt and Hollick) recovered more millipedes than a Tullgren funnel method. Since the success of a dry extraction method is dependent on the activity of the millipedes, it is the most useful method for estimating the numbers of living millipedes in soil treated with an insecticide. Those arthropods recovered in flotation methods are often dead after extraction, especially where an organic solvent is used (Raw, 1955). The conventional flotation methods using either brine (Edwards and Dennis, 1962) or magnesium sulphate (Raw, 1955) or the improved method described by Heath (1965) were discarded due to their relative complexity and the length of time needed for extraction; the Salt and Hollick flotation method often needs more than 3 days for the sorting of a single sample (Macfadyen, 1953).

3.3.1. A Simple and quick flotation technique

The basic principle that arthropod integuments are oleophilic and hydrophobic was used as the basis for separating millipedes from soil. Petroleum-spirit, not containing lead or benzene, was used as the oil phase and water for the aqueous phase, a method used for separating arthropods from foodstuffs (Murphy, 1962). In preliminary tests all stages of millipedes readily floated on water after having been coated in petroleum-spirit, and it was also effective for Collembola, Symphyla, and especially Coleoptera (wire worms and pygmy mangold beetle) which have heavily cutinized integuments. A flotation apparatus was constructed using this principle and is shown in plate 6. It consisted of a stainless-steel Ladell vessel (Ladell, 1936) supported by a metal stand over a sink. The two halves of the vessel were kept permanently clamped together, the gauze between omitted and a rubber bung fitted in the hole at the base. The vessel was positioned so that the overflow from the Ladell vessel spout poured through a fine nylon

PLATE 6. A FLOTATION APPARATUS FOR SEPARATING ARTHROPODS FROM SOIL x 1/6.


ANALAR
PETROLEUM SPIRIT

sieve (60 meshes to the inch) resting on a plastic disc cemented to a funnel. A 0.5 in. plastic-foam strip was glued round the periphery of the sieve, which, when wetted, formed a seal between it and the funnel. The funnel was heat-welded to a polythene container, the bottom of which had been removed. It was mounted upside down and held in a metal ring clamp; its screw-cap was fitted with a 2 in. length of plastic pipe and 6 in. of rubber tubing leading to the sink.

The soil sample, collected from the field in an aluminium can, was first emptied into a bucket where it was crumbled by hand and any large stones removed. The Ladell vessel was half-filled with cold water to which about 50 ml. of petroleum-spirit was added. The soil was then gently sprinkled into the vessel where the arthropods adhered to the petroleum-spirit layer and the soil and other material sank to the bottom. The water was agitated with a stirrer to form an emulsion to help release any trapped arthropods and was left for one minute for the soil particles to sink. The water level in the Ladell vessel was raised so that it carried the petroleum-spirit and arthropods over into the sieve where they were held. When the plastic vessel beneath the sieve was half full of water the clip beneath was opened. The depression in water level inside caused a suction, similar to that of a piston, which prevented the sieve from becoming clogged by organic debris and helped retrieve the petroleum-spirit lying on the sieve surface. When all the petroleum-spirit had been carried over into the sieve from the Ladell vessel, a point marked by a 'puckering' effect on the water surface, the water supply to the vessel was turned off and a 500 ml. beaker placed beneath the plastic vessel to collect the remaining water issuing from the pipe and the layer of petroleum-spirit on its surface. The recycling of the petroleum-spirit involved a loss of approximately 50% at each extraction. The contents of the sieve was then washed with a jet of water from a pipe to remove fine soil particles and transferred to a Petri dish for examination. The plug from the Ladell vessel was removed to allow the waste water and soil to flow into the sink. The time taken for the complete operation for one soil sample by an experienced operator was approximately 4-5 minutes.

For the most efficient extraction the soils must crumble easily and must not be too wet. The method was therefore unsuitable for use with soils containing clay. In practice soils were selected showing the most favourable characteristics.

3.3.2. Determination of extraction efficiency

The efficiency of the petroleum-spirit flotation method was assessed by the introduction of a known number of soil-inhabiting arthropods. Soil which had been through the extraction apparatus, was collected, oven dried, then remoistened to approximately 15-20% moisture content. Any creatures remaining in the soil would now be in an unrecognizable condition. The soil was allocated to ten aluminium cans used for sample collection. A known number of between 3 and 12 millipedes (Blaniulus guttulatus) or Collembola (Onychiurus armatus) were released into each can and mixed into the soil. After 48 hours the contents of the cans were subjected to the usual treatment involved in the operation of the flotation apparatus and the creatures collected and counted in Petri dishes in the usual way; results are recorded in table 2.

TABLE 2. EXTRACTION EFFICIENCY OF THE FLOTATION TECHNIQUE

Total creatures introduced, I and recovered, R.				Percentage recovery, %	
B.guttulatus		O.armatus		B.guttulatus	O.armatus
I	R	I	R		
66	61	66	63	92	95

These results compare well with the 81% of Collembola recovered by Kempson et al (1963) using a modified Tullgren funnel and the 70% by Heath (1965) for both modified and improved Salt and Hollick techniques. Shortage of specimens prevented the tests from being extended to the other soil-inhabiting arthropods.

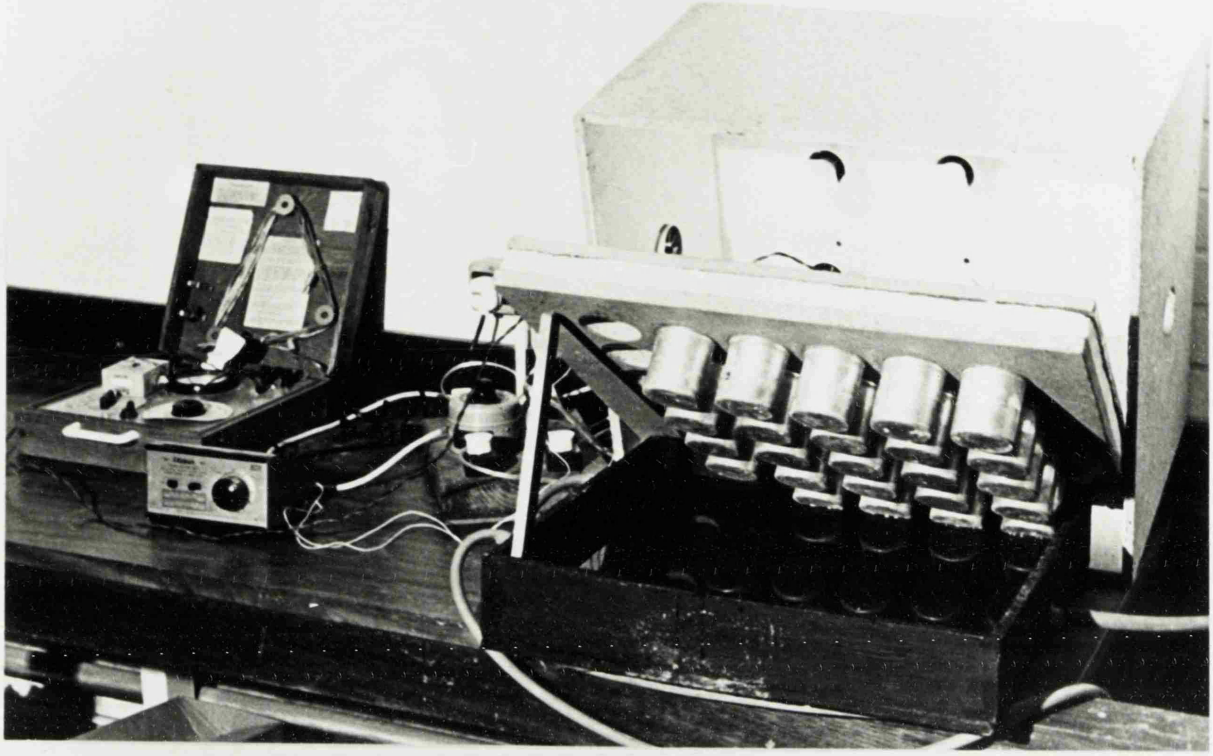
3.3.3. Other flotation methods

Heijbroek (1972) and Edwards and Dennis (1962) have extracted Onychiurus and Symphyla respectively by crumbling the soil over cold water, a method sometimes used in the present study for separation of living and dead arthropods; specific flotation methods are described fully in later sections of the thesis.

3.3.4. A prototype high-gradient canister apparatus

An apparatus, similar to that designed by Macfadyen (1961), was constructed for use in conjunction with 2.3 in. diam. x 1.9 in. deep aluminium soil auger liners: Plate 7 shows the apparatus ready to receive the soil samples. It consists of 24 (2.4 in. diam. x 3.2 in. deep) aluminium cans suspended by their screw-caps from an insulating barrier; this was formed from a lower sheet of 0.8 in. thick chipboard sandwiching a 1 in. sheet of polystyrene foam with a top sheet of 0.5 in. thick fibreboard. A 2.2 in. diam. orifice was cut in each aluminium screw-cap, each of which were glued with 'Araldite' into 2.5 in. diam. holes cut in the chipboard. Into each of the holes in the 'sandwich' was fitted aluminium sample containers 2.5 in diam. x 2.3 in. deep glued in position and supported on a 0.1 in. mesh steel gauze. Reflective aluminium baking foil was placed on the uppermost surface to reflect heat thus leaving only the sample containers uncovered. The whole structure rested on a 21 x 18 x 4 in. waterproofed water bath through which cold tap water, 2 in. deep, was circulated via an inlet and exit pipe to the sink. When in use the sample containers were covered with a hood holding a battery of four 25 w light bulbs. The light intensity (and therefore the heat produced) could be adjusted with a rheostat. A constant flow of air was ensured by fitting a small fan in the hood, operated by an integral 12 volt electric motor connected to a transformer from which its speed could be adjusted. Temperature, both at the top and bottom of the soil sample, which was placed on the gauze in each sample container, could be monitored from two thermistor probes connected to a Wheatstone bridge circuit.

PLATE 7. <u>A PROTOTYPE HIGH-GRADIENT CANNISTER APPARATUS FOR SEPARATING ARTHROPODS FROM SOIL</u>.



Soil samples were placed in the containers flush with the surface either retained in their liners, or without if the soil was easily removed. Water was placed in each of the collecting cans to a depth of 1 in. to create a high humidity beneath the sample and the hood holding the light bulbs fitted in position. The light was switched on and the intensity of the light adjusted to give a soil surface temperature of 25-30°C; the fan was set to run slowly continuously. The water flowing around the cans maintained a temperature of 10-12°C and the temperature at the bottom of the soil sample under the gauze was initially 14°C but rose later to 19°C after two weeks.

The apparatus was tested for soil samples taken at a site where millipedes, <u>Boreoiulus tenuis</u>, were numerous in the soil and where some plots had received an insecticide soil treatment. At intervals of 4, 7, 14 and 15 days of almost continuous use the contents of the collecting cans were emptied into Petri dishes and numbers of arthropods recorded. The arthropods were millipedes, <u>B.tenuis</u>; Collembola, <u>Onychiuridae</u>, <u>Isotomidae</u>, <u>Hypogastruridae</u>; and Acarina. After 15 days no more arthropods were recovered and the soil was completely dry on the under-surface of the samples. The different arthropod groups were extracted at different rates; after 4 days 50% of the total millipedes had left the soil compared with 39% of Collembola and only 25% of the Acarina (Fig. 1).

The exercise showed that blaniulid millipedes could be recovered by the dry extraction method but it must be noted that the particular species used was the smallest, slimmest blaniulid, often less than 0.6 in. long. At least one millipede was killed

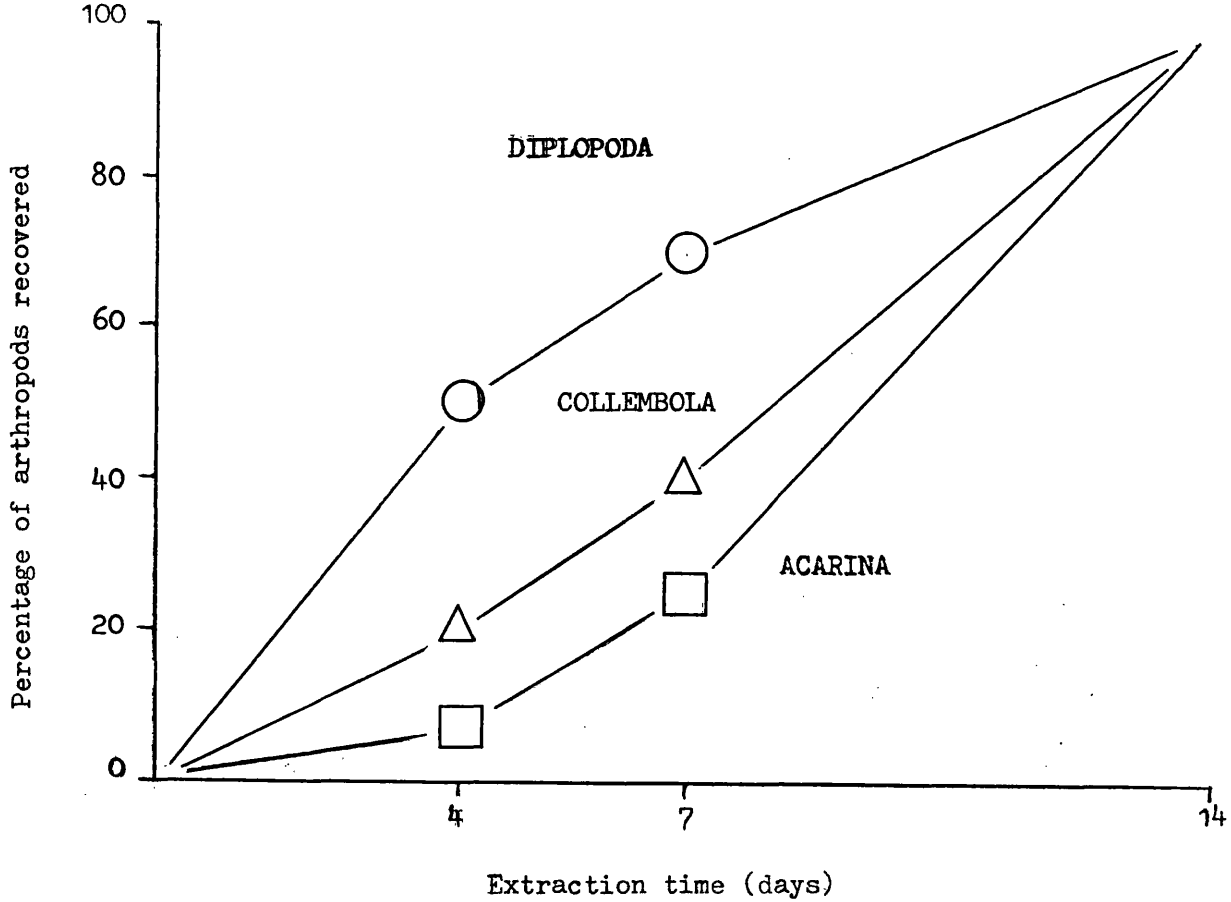


FIG. 1. A COMPARISON OF THE EXTRACTION RATE OF THREE ARTHROPOD GROUPS IN A HIGH-GRADIENT CANNISTER APPARATUS

by heat on the soil surface but examination of the dry soil, after the extraction was completed, only revealed two dried millipedes. The numbers of Collembola, which were mainly Hypogastrura spp. and Tullbergia spp. and Acarina left in the samples could not easily be assessed owing to their small size. The relative rapidity at which the millipedes left the soil samples was probably a reflection of their sensitivity to dry soil and to temperature. Light, temperature, dessication and the effect of gravity are thought to be the most important factors concerned in the movement of micro-arthropods out of samples in the Tullgren funnel method of extraction (Nev, 1962).

3.4. ASSESSMENT AND IDENTIFICATION OF ARTHROPODS

Soil-inhabiting arthropods extracted by either the flotation or dry extractors were inspected within Petri dishes in the laboratory. A small quantity (approximately 3-5 ml.) of petroleum-spirit was added to the water in which they were collected and agitated with a small paint brush to free those adhering to air bubbles at the bottom of the dish. This operation caused all the arthropods to rise and float on the water surface where they could easily be counted and identified. The different specimens were picked off with the tip of the paint brush and recorded. Identification was completed quickly because usually only Diplopoda, onychiurid Collembola and soil mites were recorded but often pygmy beetle and sometimes wireworms and Symphyla were present. In early studies, from 1970 to 1972, the different genera within the group Onychiuridae were not separated but later studies, which confirmed that *Onychiurus* spp. can be pests of sugar-beet seedlings, dictated that *Onychiurus* spp. should be recorded separately. Owing to the difficulty of separating the different species of *Onychiurus* and Acarina, samples were often sent to the British Museum (Natural History) for identification. Diplopoda were identified according to the key provided by Blower (1958); Symphyla, from Edwards (1959b).

4. SOME EFFECTS OF THE CHANGES IN SUGAR - BEET HUSBANDRY ON THE DISTRIBUTION OF SOME SOIL-INHABITING PESTS & THEIR DAMAGE TO SEEDLINGS

4.1 INTRODUCTION

The traditional method of sowing multigerm sugar-beet seed randomly and thickly in the row has now been entirely superseded by precision-drilling methods, reviewed fully by Hull & Jaggard (1971). Although the row width of 20 in. has remained the same, nearly 0.5 million acres is now sown with precision drilling equipment which, in conjunction with the use of pelleted genetic monogerm seed, enables the seed to be accurately spaced in the row. The objectives, dictated by economic pressures, aim at a decrease in the amount of labour needed to achieve the optimum plant density for maximum yield and hence the use of herbicides and planting-to-a-stand has increased. The number of seed sown on a row width of 20 in. has decreased from about 240,000 per acre in 1965 (Dunning & Winder, 1966) to about 80,000 in 1971 (Dunning, 1971).

In order to achieve the desired 30,000 plants/acre on a 20 in. row width monogerm seed may be 'planted-to-a-stand' at 5 in. spacing so that 63,000 seeds per acre should give, on average, an evenly distributed correct population (allowing for an average 60% emergence). Such a decrease in seedling populations has given rise to concern where pests may adversely affect growth before the seedlings become established. This work, concerned with the soil inhabiting pests of sugar beet seedlings, aims at answering some of the more pertinent questions arising from the changes in agronomy of sugar-beet growing: how soil-inhabiting pests are distributed in the crop and whether seedling density or herbicide usage affects the numbers of pests in the seedling root zone or the amount of damage to the roots.

4.2 MATERIALS AND METHODS

4.2.1 Sites and experiments

Sites are categorized into two groups. Firstly there are those sought for the presence of a particular pest in which to carry out field experiments and secondly those which were sampled after the infestation by a pest where additional information on numbers and distribution was required.

In the first group many fields were visited on farms in the East Anglian region in winter and early spring to estimate the size of ~~the~~ pest populations in the soil. For millipedes, which are true soil-inhabiting arthropods, the location of previous infestations when sugar beet was grown was noted. Fields at Shouldham (Norfolk), and Kettering (Northants) both had numbers in the seedbed sufficient to cause noticeable damage to the seedlings when sugar beet was last grown 4 years earlier in 1969. At Marham (Norfolk) the seedlings were damaged by millipedes in 1972 but sugar beet was grown again in 1973 and that part of the field where damage was most likely to occur again was used as the location for the experiments. Extensive preliminary sampling at Bottisham in 1971 served both to determine the presence of blaniulid millipedes and the most suitable location of experiments in the most highly infested area.

In order to ensure damage by ~~a~~ pests such as pygmy beetle, *Atomaria linearis*, which overwinters on the fresh growth of old beet crowns in the soil, sugar beet was grown for a second consecutive year at six sites: Boston (Lincs.) and Welney (Norfolk) in 1971; Magdalen (Norfolk), Stokesby (Norfolk) and Benwick (Norfolk) in 1972 and Marham (Norfolk) in 1973. In the previous year's crop pygmy beetle had been the main pest but millipedes, *Brachydesmus superus* and *Blaniulus guttulatus* were also present at Welney and

Magdalen sites respectively.

Most sites could be assumed to have small populations of Collembola (Onychiurus spp.). At a site at Stalham in 1974 a field was chosen opposite one, where in 1972, high numbers had been responsible for poor seedling emergence and no other pests had been present in significant numbers.

At Littleport in 1973, a site where Symphyla had caused much damage when sugar beet was last grown, experiments were placed on that part of the field where most damage had occurred. Experiments were also carried out at Broom's Barn (Suffolk) where problems attributable to soil-inhabiting pests were rare.

Table 3 shows the type of experiment and design details at sites where soil-inhabiting pests had been found. Randomised block experiments investigated whether different seed spacings or herbicide treatments affect the numbers of pests in the root zone of sugar-beet seedlings and their distribution in the crop, particularly numbers in the row and between rows. Except at one site, three seed spacings were compared. Pelleted sugar-beet seed, cultivar Amono, was used in 1971 and 1972, Monotri or Amono in 1973 and Monotri in 1974; they were all pelleted after steeping in ethyl mercury phosphate (EMP) fungicide but were not treated with an insecticide. Seed was sown with a 'Stanhay' 5-row, tractor-mounted seed drill. Initially chosen as a comparison, were spacings of 1.5 in., 3 in. and 9 in. to cover the spectrum of commercial sugar-beet seed spacings but in later experiments, from 1972-1974, the 3 in. spacing was replaced by one of 4.5 in. Plots were usually 5 rows wide in 1971-1973 with each row 20 in. apart.

TABLE 3. DETAILS OF SEED SPACING AND HERBICIDE EXPERIMENTS 1971-1974

Year	Site	Number of treatments spacing/herbicide (in.)		Seed cultivar	Layout. Treatment x blocks	Rows/ plot	Plot length (ft.)
1971	Bottisham	1.5, 3, 9	±	'Amono'	6 x 4	5	43
	Broom's Barn	"	"	"	6 x 4	5	50
	Welney	"	"	"	6 x 4	5	50
1972	Benwick	1.5, 4.5, 9	±	'Amono'	6 x 4	5	50
	Broom's Barn	"	"	"	6 x 4	10	65.3
	Magdalen	"	"	"	6 x 4	5	100
	Melbourn	"	"	"	6 x 4	5	100
	Shouldham	"	"	"	6 x 4	5	50
1973	Black Bank	1.5, 3, 4.5	+	'Monotri'	3 x 5	4	56
	Broom's Barn	1.5, 4.5, 9	±	'Amono'	6 x 4	10	50
	Kettering	"	+	"	3 x 6	5	66
	Littleport	"	"	'Monotri'	3 x 4	5	50
	Marham	"	±	"	6 x 4	5	58
	Stokesby	3.8, 7.5	+	"	2 x 4	6	48
1974	Bottisham	1.5, 4.5, 9	+	'Monotri'	3 x18	5	70
	Gedney	"	"	"	3 x 9	5	50
	Stalham	"	"	"	3 x18	5	63

Since it was important that row width conformed to that used by the grower, that at Stokesby was 18 in. At Black Bank (Norfolk) and Stokesby plots were 4 and 6 rows wide respectively; at these two sites seed spacings were also anomalous at 3 in. and 4.5 in. and 3.8 in. and 7.5 in. respectively. A pre-emergence herbicide treatment was included in seed spacing experiments in 1971 and 1972 and at Broom's Barn and Marham in 1973 making a total of 6 treatments: 3 spacings, plus and minus herbicide. The inter-row spaces of plots where rows received herbicide were sprayed with herbicide from a knapsack sprayer shortly after drilling. Some experiments in 1973 and all in 1974 received only a band-applied herbicide, which is standard commercial practice, with none between the rows. On all experiments, at the time when the seedlings were large enough for singling, any inter-row weeds were removed by a tractor-mounted hoe. Single-row plots were used in 1974 to achieve more extensive replication at Bottisham, Gedney, (Norfolk) and Stalham. Each of the three centre drill units sowed seed at either 1.5, 4.5 or 9 in. spacing whilst the two outer rows were sown with seed at 3 in., to be used as discard rows; all rows were band-sprayed with herbicide at drilling.

Sites in the second category were visited from 1970-1972 following reports by B.S.C. fieldmen that soil-inhabiting pests were causing damage to commercial crops. Individual fields having a predominance of one of the major pests were sampled, the numbers of the pest involved ascertained and damage to the seedlings inspected.

4.2.2 Soil sampling and extraction of pests from the soil

The soil was sampled at all sites where pests were present. Preliminary sampling in bare soil provided a basis for the suitability of a site for an experiment.

Table 4 shows details of sites, when samples were taken, and number and size of sample units in preliminary sampling. Information on the movements of pests in the soil was obtained by taking samples at different depths; also, by sampling on successive dates variations in the population size could be monitored. In 1971 only one site, Bottisham (Cambs.), was investigated for blaniulid millipedes; pygmy beetle was the main target in this year and sites at Boston and Welney were investigated where sugar beet was to be grown for the second year in succession. Extensive preliminary sampling was done only in 1971 and not extended to the future programme. Unnecessary effort and time was not expended by extensive preliminary sampling at sites where the likelihood of suitable infestations by pests was high. From 1972 to 1974 a single preliminary sample was taken from the top 3.9 in. of the soil as near to the time of drilling as possible to allow for the upward migration (displacement) of the maximum number of creatures.

Seed spacing and herbicide experiments were located where suitable numbers of pests were present in the seedbed. Soil samples were always taken both in the row and midway between rows at the time of drilling the crop to estimate initial seedbed pest populations. Table 5 shows the experimental design, number of treatments, size and number of rows per plot at the different sites. To monitor the upward displacement of soil-inhabiting pests into the seedbed soil cores were taken at random midway between the rows shortly after the seedling cotyledons had emerged above the soil surface. Unlike those taken at other sites, samples

TABLE 4. PRELIMINARY SOIL SAMPLING AT PEST-INFESTED SITES

Year	Site	Date	Sampling depth (in.)	Number and size of units, diam. x depth (in.)
1971	Boston	25.2.71	(0 - 6	20 2x6
	"	"	(6 -12	20 2x6
"	"	20.4.71	(0 - 3.9	16 2.3x3.9
			(4.7- 8.7	16 2.3x3.9
"	Welney	29.1.71	(0 - 6	10 2x6
			(6 -12	10 2x6
"	"	19.2.71	(0 - 6	10 2x6
			(6 -12	10 2x6
"	"	19.3.71	(0 - 6	10 2x6
			(6 -12	10 2x6
"	"	13.4.71	(0 - 3.9	16 2.3x3.9
			(4.7-8.7	16 2.3x3.9
"	Bottisham	22.2.71	(0 - 6	5 2x6
			(6 -12	5 2x6
			(12 -18	5 2x6
"	"	4.3.71	(0 - 6	50 2x6
			(6- 12	50 2x6
"	"	14.4.71	(0-3.9	10 2.3x3.9
			(4.7-8.7	10 2.3x3.9

TABLE 5 SEED SPACING AND HERBICIDE EXPERIMENTS 1971-1974; SOIL SAMPLING RECORD

	Site	'At drilling' soil sample Date	Number of units	Size of units diam. x depth (in.)
1971	Boston	20.4	16	2.3 x 3.9
	Bottisham	13.4	24	"
	Broom's Barn	19.4	24	"
	Welney	13.4	24	"
1972	Benwick	27.3	24	2.3 x 3.9
	Broom's Barn	none		
	Magdalen	30.3	24	"
	Melbourn	17.4	24	"
	Shouldham	20.4	48	"
1973	Black Bank	11.4	24	2.3 x 3.9
	Broom's Barn	26.3	24	"
	Kettering	17.3	24	"
	Littleport	30.3	24	"
	Marham	12.4	24	"
	Stokesby	23.3	24	"
1974	Bottisham	8.4	12	8 x (1 x 3.9)
	Gedney	3.4	17	"
	Stalham	11.4	12	"

Continued on P. 41

TABLE 5 (CONTINUED)

Year	Site	Date	Post emergence soil sample Number of units 'in row'	between rows	Size of units, diam. x depth, (in.)
1971	Boston	none			
	Bottisham	none			
	Broom's Barn	27.5	48	0	2.3 x 3.9
	Welney	17.5	24	24	"
1972	Benwick	none			
	Broom's Barn	11.5	24	24	2.3 x 3.9
	Magdalen	5.5	24	24	"
	Melbourn	none			
	Shouldham (	a 19.5	24	24	"
	(	b 6.6	48	24	10 x(1 x 3.9)
1973	Black Bank	4.6	30	0	8 x(1 x 3.9)
	Broom's Barn	17.4	24	24	5 x(1 x 3.9)
	Kettering (	a 25.4	24	24	8 x(1 x 3.9)
	(	b 14.5	27	27	"
	(	c 30.5	24	0	"
	Littleport (	a 2.5	24	24	"
	(	b 24.5	24	0	"
	Marham (	a 8.5	12	12	"
	(	b 17.5	18	18	"
	(	c 6.6	18	0	"
	Stokesby	22.5	16	0	"
1974	Bottisham	14.5	36	0	8 x(1 x 3.9)
	Gedney	none			
	Stalham	6.5	36	0	"

from Broom's Barn in 1973 were taken before seedling emergence; some cores contained a seedling and some did not but the number of creatures in and midway between rows could be compared. The latest samples at most sites were taken shortly before, or at the time the seedlings were singled (usually at the 4 or 6 true leaf stage).

To compare the effect of seedling spacing on pest distribution in the row, cores were taken centred over and including seedlings only when they were at the specified spacing in the row. Germination is usually in the region of 60 or 70%, implying that, on average, one third of the seedlings fail to emerge. However, there are short lengths of row where 100% germination is apparent and the seedlings are correctly spaced. Conversely there are short lengths of row where seedlings are more sparse, the seeds having germinated poorly and these areas were avoided. Cores were only taken over seedlings which were bounded on either side by a series of uniformly spaced seedlings. The term 'seed spacing' can therefore be replaced by 'seedling spacing'.

The sample unit used at all sites consisted of a soil core taken with an auger; the core size varied according to the size of auger used. In 1970, samples were taken with an auger 2 in. diameter and 6 in. deep but in 1971 one 2.3 in. cutting a core 3.9 in. deep was used. For samples taken on 19 May at Shouldham a half-round auger, 1 in. diameter cutting a core 3.9 in. deep was used. Ten such cores were bulked together in the field and 2 such bulked samples were taken within the row from each plot. Subsequent samples in 1973 and 1974 were taken with another auger designed to cut 3.9 in. deep cores 1 in. diameter. As at Shouldham, the individual cores from each plot were bulked together to form a sample unit which usually consisted of 8 cores - this was the maximum number which would fit conveniently into each screw-top aluminium can.

The number of sample units taken affects the accuracy that can be expected. In preliminary soil sampling and the 'at drilling' samples the emphasis was mostly upon expediency since many sites had to be sampled within a short period of time. For the 'post-emergence' sampling as many sample units as could be conveniently handled were taken consistent with accuracy desired. In 1972 it became apparent, from the variability of previous samples, that it was necessary to sample more seedlings which was done by bulking individual cores whilst maintaining the number of units as high as practicable. In 1971 and 1972 samples were usually taken from both in the row and between the rows but from 1973 to 1974 the emphasis changed to more extensive sampling in the row.

Details of the sampling at sites which were visited after the pest attack had occurred are shown in Table 6. In 1970 soil samples were taken in the row between seedlings in addition to the other sampling locations.

Great care was taken when handling samples to avoid crushing the soil. For this reason soil cores, whether as individual units or bulked, were transferred immediately from the auger in the field to a screw-top aluminium can which afforded protection from desiccation and in handling and storage. In the laboratory the cans were stored until the creatures could be

TABLE 6 SOIL SAMPLING RECORD AT SITES WHERE SOIL-INHABITING PESTS WERE CAUSING DAMAGE TO SUGAR - BEET SEEDLINGS

Year	Site	Date	Sampling depth (in.)	Number of sample units			Unit size (in.)
				In row	In row between seedlings	Between rows	
1970	March	13.5.70	0 - 6	20	20	20	2 x 6
	Bottisham	a12.6.70	0 - 6	5	5	0	2 x 6
		b12.6.70	0 - 6	5	5	0	2 x 6
	N.Nynn	12.5.70	0 - 6	25	-	25	2 x 6
1971	Bottisham	4.6.71	0 - 3.9	16	-	16	2.3 x 3.9
1972	Stalham	28.4.72	0 - 3.9	12	-	12	2.3 x 3.9

extracted, usually for a period of up to one week at $5^{\circ}C$. Occasionally the contents of the cans were transferred to polythene bags to be frozen at $-15^{\circ}C$. This method of storing could be used only on those samples where pygmy beetle were to be recorded since they were the only creatures that remained recognizable after thawing.

The arthropods were extracted by the Petroleum-spirit flotation method described in section 3.3.1.

4.2.3 Seedling assessment (establishment, root damage and vigour)

Seedling establishment relates the number of seeds sown to the number of seedlings produced and it is the most useful parameter of practical value when assessing damage to the crop at this stage. The relationship investigated was between both the initial pest numbers in the seedbed and numbers of pests per seedling root with seedling establishment. At all sites, except at March in 1970, the number of seedlings per unit length of row was counted. The assessment of damage to the roots of seedlings was done at various sites from 1971 to 1974, the details of which are shown in Table 7; a scoring method was used with a scale of 0 to 5. Sites in 1971 and 1972 included a herbicide treatment in addition to the three seed spacings. Seedlings were removed at random from the plots except at Shouldham where a length of row containing 20 adjacent seedlings was removed from a single row. In 1974 an additional seedling sample was obtained by removing the seedlings from the soil cores taken to measure the number of pests per seedling root, the sample comprised only 8 seedlings per plot but the number of plots sampled was increased. The seedlings were carefully removed

TABLE 7 SEEDLING SAMPLING FOR ROOT DAMAGE AND DRY WEIGHT ASSESSMENTS ON SEED SPACING AND HERBICIDE EXPERIMENTS 1971-1974

Site Sampling date	Bottisham 2.6.71	Broom's Barn 26.5.71	Broom's Barn 3.6.72	Magdalen 5.6.72	Melbourn 18.5.72	Shouldham 1.6.72	Bottisham (i) 8.5.74 (ii) 10.5.74	Stalham (i) 6.5.74 (ii) 21.5.74
Treatments								
a 1.5 in. spacing	a	a	a	a	a	a	-	-
b " " " + herbicide	b	b	b	b	b	b	b	b
c 3 in. spacing	c	c	-	-	-	-	-	-
d " " + herbicide	d	d	-	-	-	-	d	d
e 4.5 in. spacing	-	-	e	e	e	e	-	-
f " " + herbicide	-	-	f	f	f	f	f	f
g 9 in. spacing	g	g	g	g	g	g	-	-
h " " + herbicide	h	h	h	h	h	h	h	h
Number of seedlings sampled per plot	50	30	50	23-26	24	20	(i) 8 (ii) 24-28	(i) 8 (ii) 24-27
Number of plots sampled	12	24	24	24	24	24	(i) 36 (ii) 27	(i) 36 (ii) 27

from the soil with a trowel taking care to include soil surrounding the root system and those from each plot bulked together in a polythene bag. In the laboratory the soil adhering to the root was removed with water and the roots inspected for lesions. The score given for damage depended both on the number and extensiveness of the lesions and was not dependent on the size of the root system as the seedlings would vary in size. A score of 0 was given where there was no discernable damage by soil pests and a score of 5 given when damage was extensive. In 1971, on the seed spacing experiment at Broom's Barn, the damage was assessed by counting the number of individual 'bites' made by pygmy beetle on the root.

Seedling vigour is a subjective measure of the size of the seedlings and quality of growth. It is important since it relates to the tolerance of seedlings to damage. At two sites, Bottisham in 1971 and at Magdalen in 1972, the seedlings in each plot were given a score (0 = poor, weak growth; 5 = very large, sturdy seedlings). The assessment was based on the relative sizes of the seedlings from the soil.

The dry weight of seedlings is a measure of the amount of tissue present and is a convenient method of comparing seedling growth on different treatments.

The seedlings collected from all sites shown in Table 7, with the exception of Magdalen, were oven-dried at 105°C until no further weight loss was recorded.

4.3 RESULTS

4.3.1 Pest populations in the soil and relationship to seedling damage

4.3.1.1 Preliminary sampling at pest sites.

In this section are included the results of soil sampling in 1971 before the crop was sown where information was required on pest numbers in the soil. Table 8 shows how the numbers of millipedes varied at the different depths in the soil at Bottisham on three successive sampling dates prior to drilling the crop. The first sample taken on 22nd February in an area 120 ft. x 60 ft. at the bottom of the intended trial area indicated the presence of blaniulid millipedes with most below 6 in. In March, 50 samples were taken over the whole field on a 10 x 5 grid system to find the best location for a trial site and most millipedes were again found below 6 in. with a concentration in one half of the field, hence the low overall population estimate of only 1.8 million per acre. Having defined the trial area, samples taken in April indicated 4.1 million blaniulids per acre; again most were found in the deepest samples taken to a depth of 7.8 in.

Soil samples at two depths at both Boston and Welney taken on diagonal transects across the two fields showed that as many pigmy beetle were present in the deeper soil cores as in the top cores. The apparent pygmy beetle population at Boston fell from 1.2 to 0.7 million from February to April; more than half of the beetles in the early sample were not intact after extraction. The extrapolation of numbers of pygmy beetle per core to the population per acre was based on the area of the soil core taken; only beetles which were intact were included in the final population estimate since they were probably live before extraction. At Welney the

Table 8 PRELIMINARY SOIL SAMPLING AT BOTTISHAM (1971)

Sampling (2 in. or 2.3 in. diam. cores). Date	Depth(in.)	Millipedes per sample unit per stratum	Millipede density per acre, for all strata (millions)
22.2.71	0 - 6	0.2	4.0
"	6 - 12	2.0	
"	12 - 18	1.8	
4.3.71	0 - 6	0.3	1.8
"	6 - 12	0.6	
14.4.71	0 - 3.9	0.8	4.1
	4.7- 7.8	2.0	

population in the top 12 in. varied from 1.8 to 3.0 million when 7.8 in. depth of soil was sampled (Table 9).

Of those beetles recovered in January and February at Welney, 58% were distinctly light brown or golden coloured. Both at Boston and Welney, Brachydesmus millipedes were common in early samples as adults but in April egg clusters only were recovered. Onychiurid Collembola were common at both Boston and Welney and usually most numerous in the deeper samples. Samples included the genus Onychiurus but it was not separated from the other genera - Collembola were the most common single group at both sites; at Bottisham they were also common but not recorded.

4.3.1.2. Resident pest populations and subsequent seedling establishment at chosen sites.

The seedbed population estimates of pygmy beetle and blaniulid millipedes were calculated from the mean number found in soil samples taken at the time of drilling the crop. The soil samples were composed of many cores 3.9 in. deep and 2.3 in. diameter which, on an area basis, was equivalent to 1.47 million to one acre; in 1974 the equivalent sample was composed of cores 1 in. diameter, equivalent to 4 million to one acre. In order to obtain a representative figure for seedling establishment over the whole trial area where the soil samples were collected a mean was taken of plots, receiving no insecticides, which were distributed at random in the various experiments over the trial area. Seedling establishment was plotted against estimated numbers of pygmy beetle and blaniulid millipedes in the top 3.9 in. of the seedbed (Fig.2). Poor seedling establishments, of less than 10%, occurred when between 0.2 and 1.8 million pygmy beetle were found in the seedbed at three

TABLE 9. PRELIMINARY SOIL SAMPLING AT BOSTON & WELNEY (1971)

BOSTON		Number of pests					
		per sample unit			per acre (million) to maximum depth		
Sampling Date	Depth (in.)	Pygmy beetle	*Milli-pedes	Onychiurid collembola	Pygmy beetle	*Milli-pedes	Onychiurid collembola
25.2.71	0 - 6	0.3	0.8*	4.4	1.2	3.2	9.0
	6 - 12	0.3	0.8*	0.1			
20.4.71	0 -3.9	0.3	0.3	2.4	0.7	1.9	15.4
	4.7-7.8	0.2	1.1	8.0			
WELNEY							
29.1.71	0 - 6	1.2	0.6	1.6	3.0	3.6	13.8
	6 - 12	1.1	0.4	5.3			
19.2.71	0 - 6	0.4	0.2	4.4	1.8	0.4	25.2
	6 - 12	0.5	0	8.2			
19.3.71	0 - 6	0.2	0.2	1.6	2.4	0.8	15.0
	6 - 12	1.0	0.2	5.9			
13.4.71	0 -3.9	1.3	0	3.1	3.5	0	16.3
	4.7-7.8	1.1	0	7.9			

* Mainly Brachydesmus superus

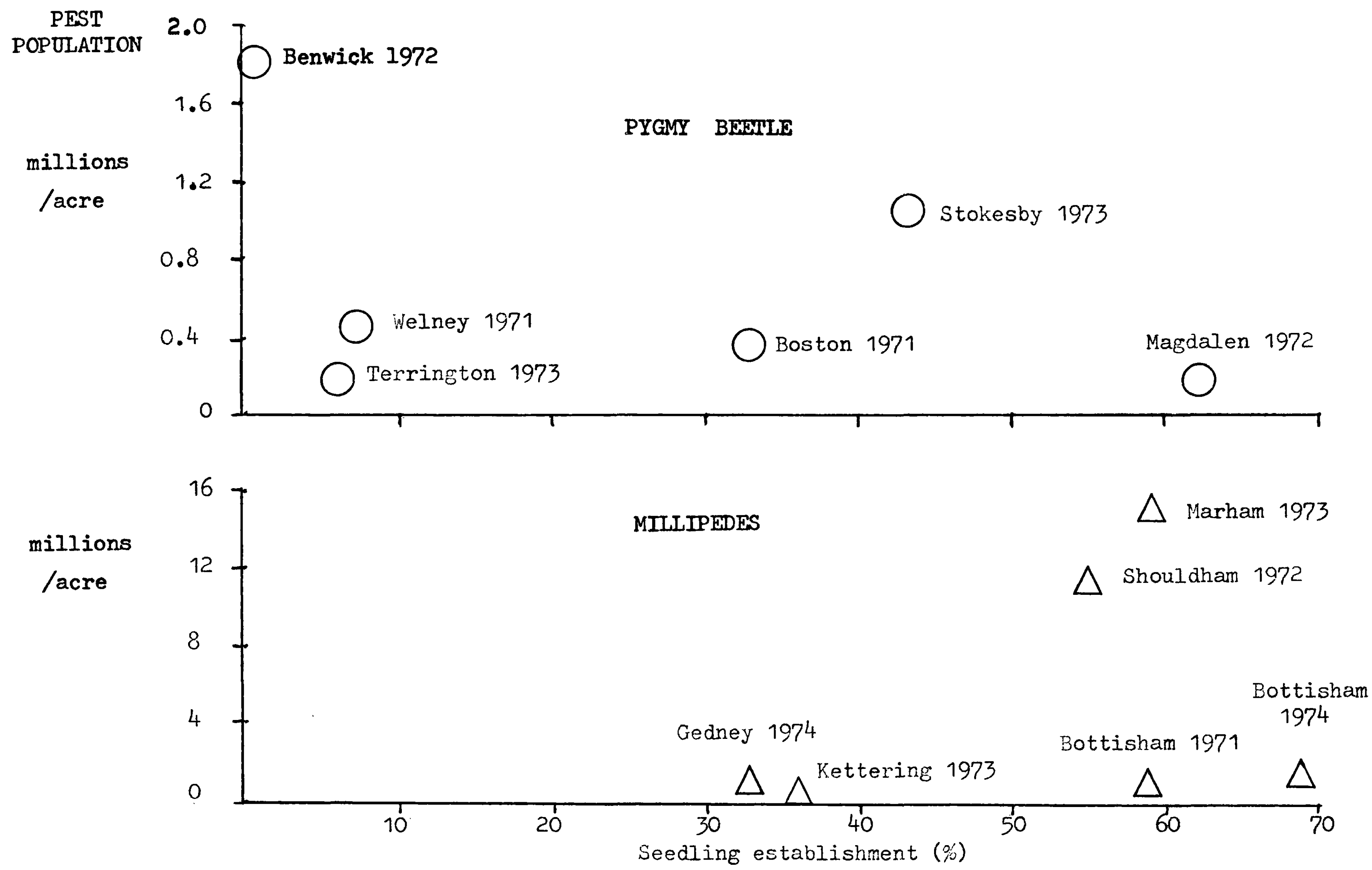


FIG. 2. ESTIMATED POPULATIONS OF PESTS IN THE SEEDBED RELATED TO SEEDLING ESTABLISHMENT.

sites, but at one, Terrington, poor establishment was probably attributable to the consolidation of the topsoil which prevented emergence of some seedlings. At Benwick the crop was redrilled.

Populations of millipedes in the seedbed were more than 10 million/acre at two sites but seedling establishment remained high. At Kettering, though very few were found at the time of drilling, damage resulted in only a 36% establishment. At Gedney an especially dry seedbed prevented germination of many seed.

4.3.1.3. Seedling infestations and seedling establishment at chosen sites

The number of pests per seedling root were found from soil samples taken at the time damage was occurring, usually from mid-May until early June. The cores were taken from experimental plots on either soil compaction experiments (Welney, Bottisham and Boston, 1971), seed spacing experiments (Broom's Barn 1971; Magdalen, Shouldham, 1972; Stokesby, Kettèring, Marham, 1973; control plots of a seed treatment experiment at Terrington 1973 and from a field without experiments at a site at March (Cambs.) in 1970). Except at the latter, no insecticide treatment had been used in the sampling area. At the March site pygmy beetle had caused such extensive damage to the seedlings of a commercial crop (treated with 0.2% dieldrin in the pellet) that, in order to avoid a crop failure, the grower re-drilled the beet; soil samples were taken immediately before re-drilling. Seedlings in all samples were at the 2-4 true leaf stage. Most data was collected for pygmy beetle and for millipedes. Seedling establishment was plotted against numbers of either pygmy beetle or blaniulid millipedes in the seedling root zone (Fig.3). For both pests the

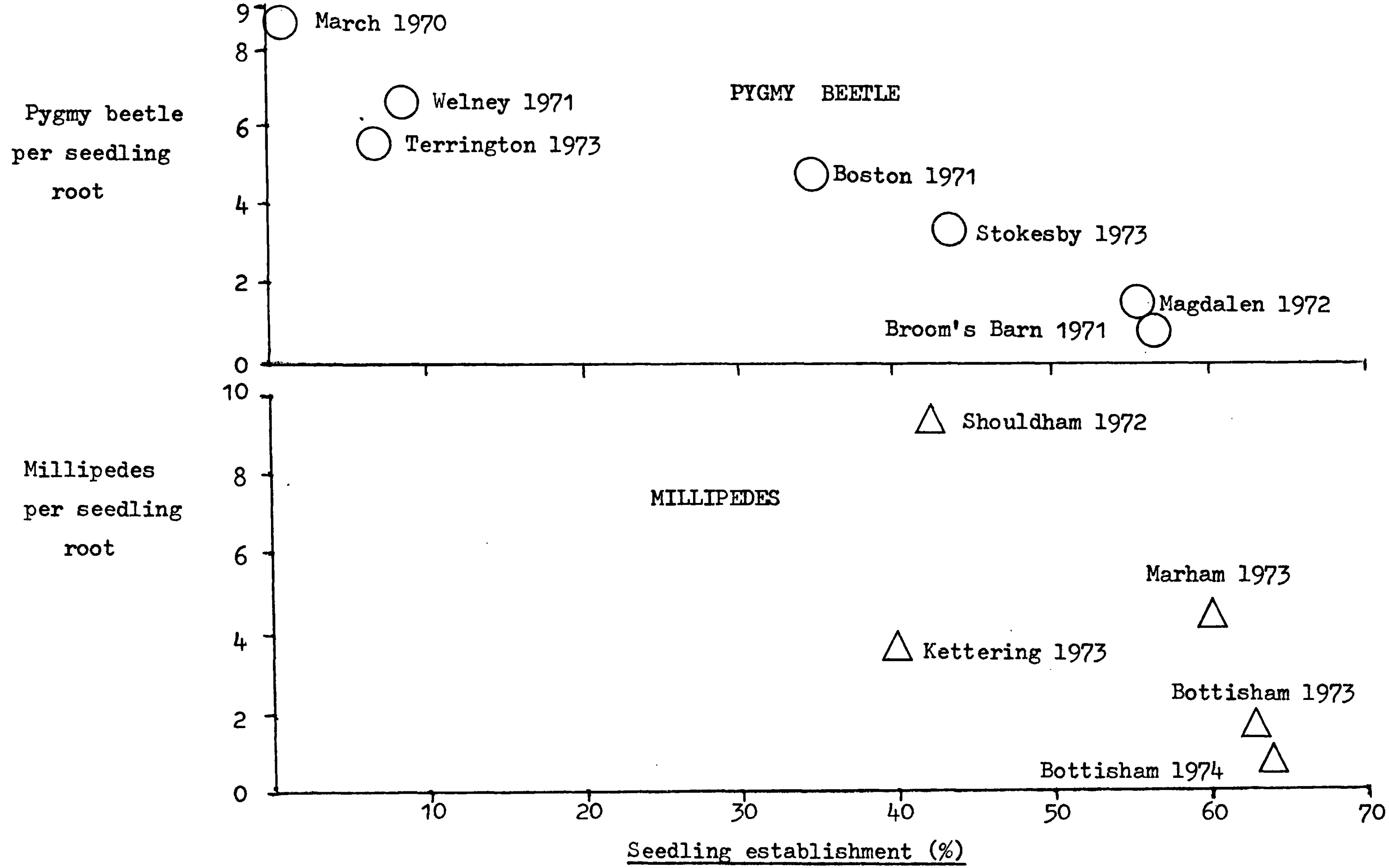


FIG. 3. ESTIMATED NUMBERS OF PESTS PER SEEDLING ROOT ZONE RELATED TO SEEDLING ESTABLISHMENT

percentage of seedlings that became established increased as the number of pests in the root zone decreased. At no sites with millipede infestations, was seedling loss particularly severe overall; seedling establishment could be up to 40% when a mean of nine millipedes were recorded per seedling root. At three sites establishment approximated to the national average of 60-65% though millipedes were active around the seedlings. At sites infested with pygmy beetle, a mean of between five and nine beetles per root was sufficient for only a 10% seedling survival; seedling establishment increased progressively as fewer beetles were found in the root zone.

4.3.1.4. Pest infestations at other sites.

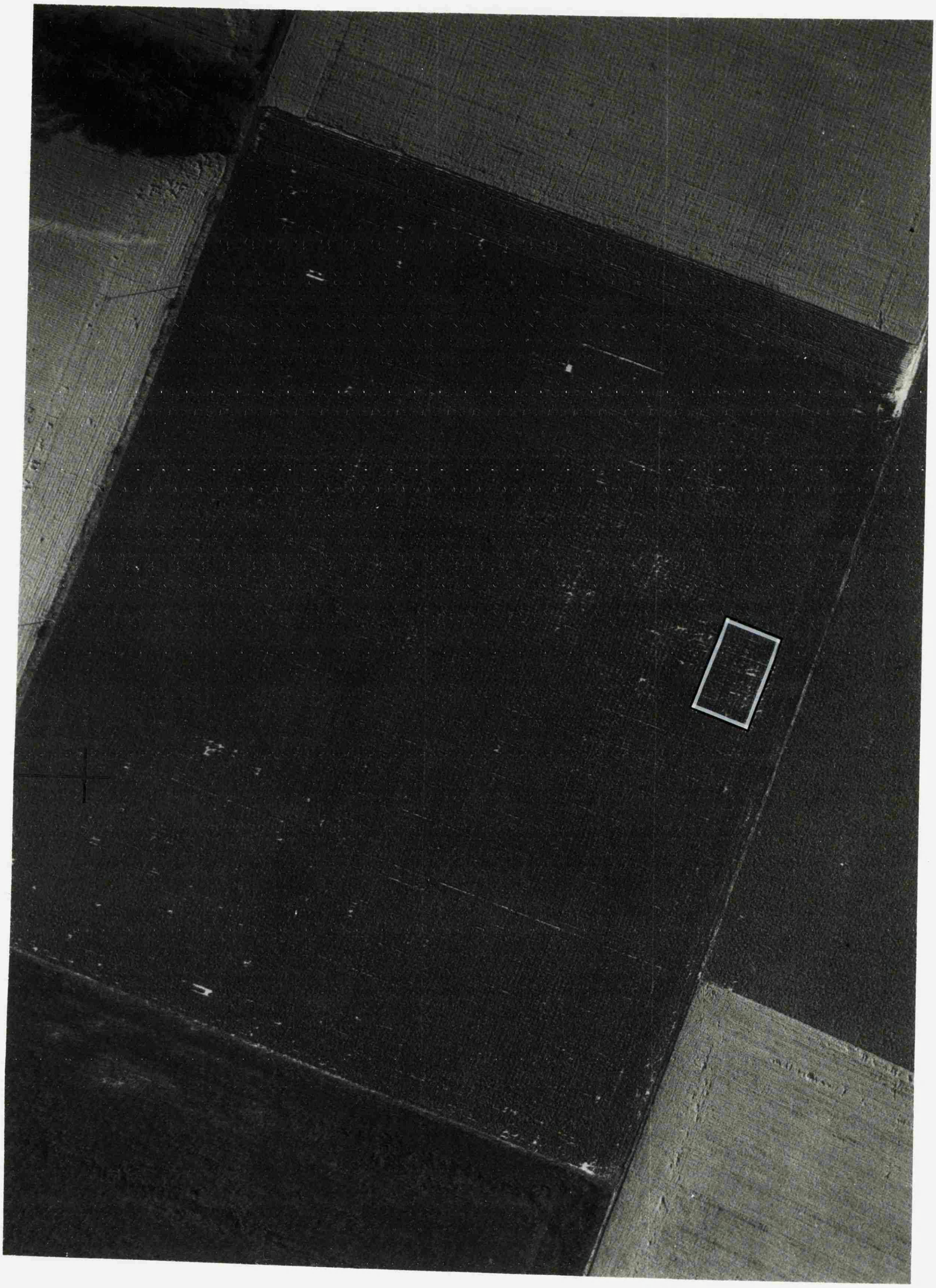
The result of taking random soil samples in pest-infested areas is shown in Table 10. At March (Cambs.) 1970, where the damage was so severe that the crop had to be redrilled, the seedlings were showing nutrient deficiency symptoms and roots or hypocotyls just at or below soil surface were sometimes completely excised; no other pests except pygmy beetle were recovered. At Bottisham (Cambs.), 1970, the millipedes, mainly Boreoiulus tenuis but some Blaniulus guttulatus, were mostly found in the area of the field where the seedlings were damaged and seedling growth poor (Sample a). A few pygmy beetles, Onychiuridae and Symphyla, were also recovered but only in very small numbers; few millipedes or other pests were found where seedling growth was vigorous. On the same farm, but on a different field, in 1971, on an area adjacent to that where experiments were sited, the same millipede species was causing damage in a small patch of sugar beet near to the headland. Plate 8 is an aerial photograph of the field taken in mid-August; the area where the seedlings had been damaged

TABLE 10. THE DISTRIBUTION OF SOIL-INHABITING PESTS AT SITES WHERE THEY WERE CAUSING DAMAGE TO SUGAR-BEET SEEDLING ROOTS

Site	Date	Main pest	Mean number of pests per sample unit		
			In row around the seedling root	In row between seedlings	Between rows
March (Cambs.)	13.5.70	Pygmy Beetle	8.6	0.2	0.1
Bottisham (Cambs.)	12.6.70	Millipedes a.	44.0	2.0	-
		b.	1.2	0	-
N.Lynn (Norfolk)	12.5.70	Symphyla	2.6	-	0.4
Bottisham (Cambs.)	4.6.71	Millipedes	49.3	-	2.3
Stalham (Norfolk)	28.4.72	Collembola	18.1	-	6.4

PLATE 8. AERIAL PHOTOGRAPH OF THE FIELD AT BOTTISHAM (CAMBS.) IN 1971, SHOWING LOCATION OF TRIAL AREA AND A PATCH WHERE MILLIPEDES HAD CAUSED LOCAL LOSS OF SEEDLINGS.

(See text for explanation of plate)



(outlined in white in the plate) shows a less-dark foliage cover than the rest of the crop due to the earlier loss of seedlings. The high numbers of blaniulid millipedes per sugar-beet seedling root (a mean figure of 49) compares with the corresponding figure of 44 per root at Bottisham in the previous year in a similar localised patch of infestation. At North Lynn (Norfolk) symphylids caused a similar but more extensive pattern of damage; seedlings showed the characteristic stunted lateral roots where the symphylids had been feeding which caused many seedlings to wilt and ultimately to die. At Stalham concentrates of *Onychiurus* spp. were found around seedling roots in the area showing seedling loss. An average of 18 per seedling was recorded but at the time they were not considered to have caused the primary damage.

4.3.2. Distribution of some soil-inhabiting pests in the crop

4.3.2.1. Distribution in time.

Soil samples taken at three times and covering the period from late April to early June at Kettering and Marham in 1973 showed that the millipede infestation rose to a peak in mid-May and declined in June. Not only did the size of the population in the row increase (Fig.4) but also the proportion of individuals in the row increased from the time the crop was sown until the last samples when up to 98% of the millipedes were found around the seedling roots (Fig.5). An exactly similar pattern was evident in samples taken at Shouldham in 1972 when the crop was sown later.

Pygmy beetle, unlike millipedes, were not numerous in the early samples taken in the late April-early May period and were often absent. Later samples in May and June at Kettering and Marham show a gradual increase in the size of infestation, whilst at Littleport in the same year, numbers increased sharply from a mean number of 0.5 to over 38 per sample unit (a mean of almost 5 beetles per seedling) during May (Fig.6).

At Littleport, Symphyla were also causing damage to roots. They averaged 3.2 and 1.0 per sample unit in early and late May samples respectively.

Onychiurid Collembola were not recorded in samples taken on successive dates in 1972 and 1973 but there was evidence from observations on seedling damage at Broom's Barn in 1973 that they can be active in the seedbed in March and April before other pests such as millipedes or pygmy beetle.

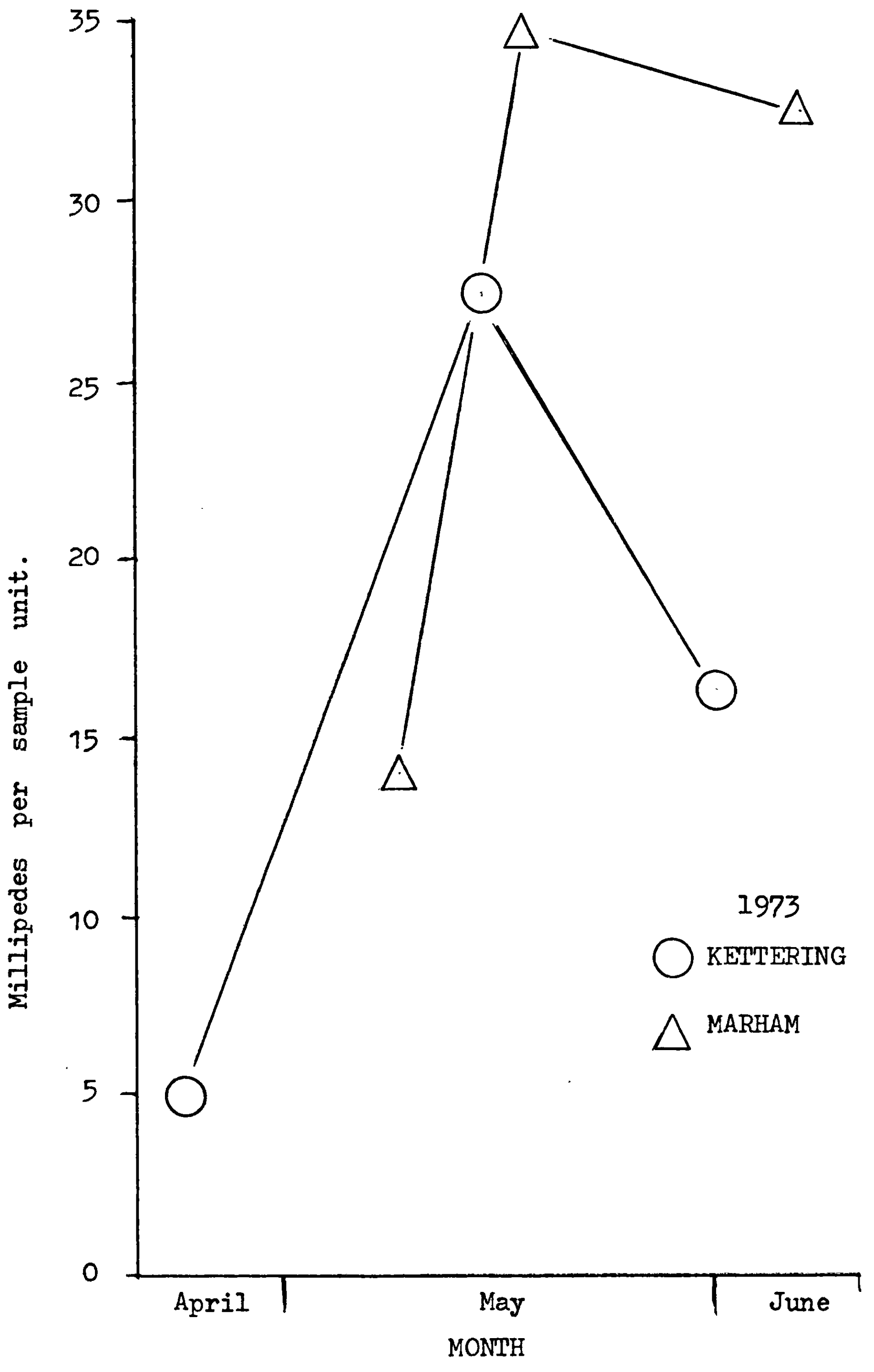


FIG.4. NUMBERS OF MILLIPEDES IN THE ROOT ZONE OF SEEDLINGS ON SUCCESSIVE SAMPLING DATES AT TWO SITES.

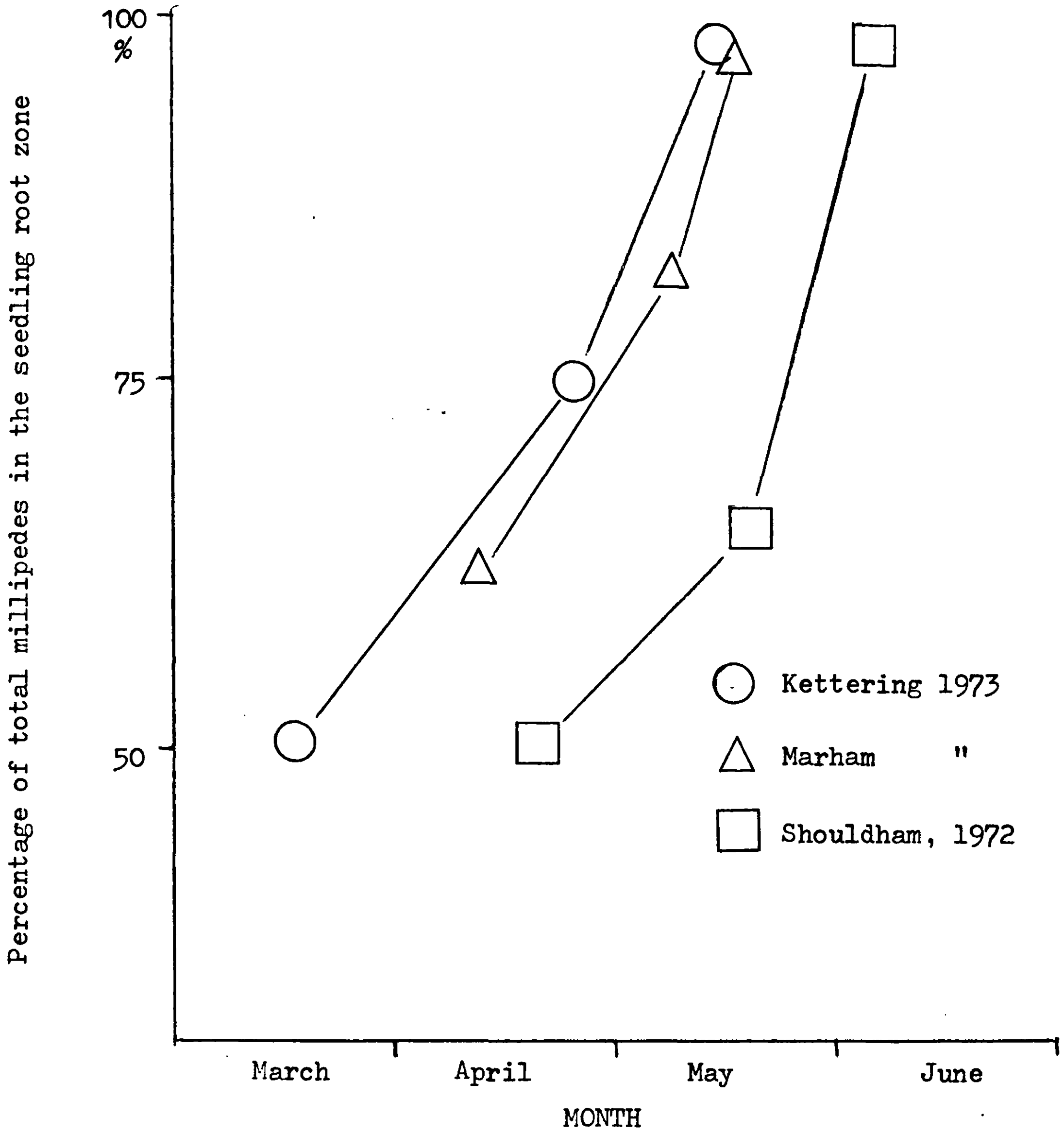


FIG. 5. THE PROPORTION OF THE TOTAL MILLIPEDES IN SOIL SAMPLES THAT ARE FOUND IN SEEDLING ROOT ZONE ON THREE SUCCESSIVE SAMPLING DATES.

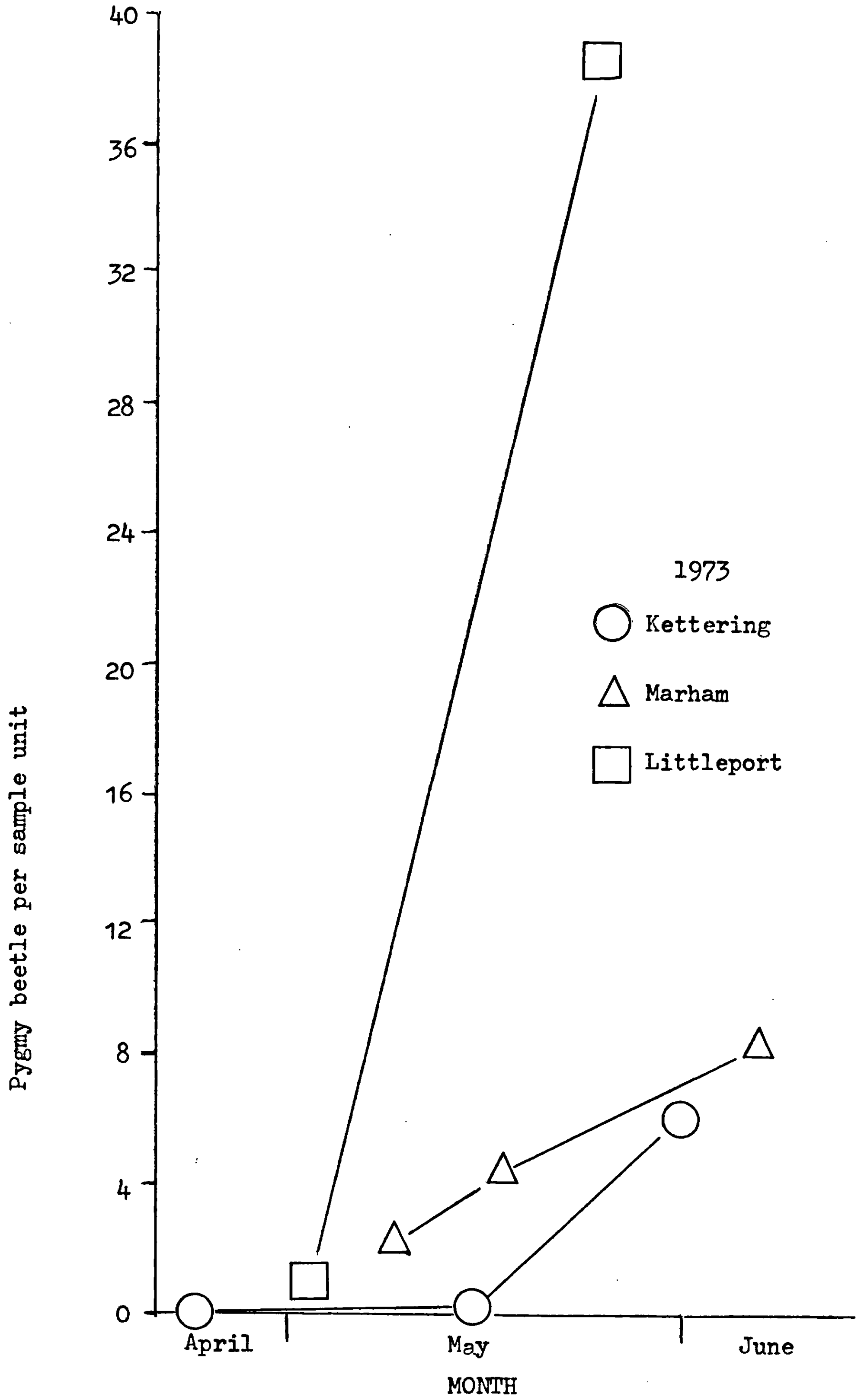


FIG. 6. NUMBERS OF PYGMY BEETLE IN THE ROOT ZONE OF SEEDLINGS ON SUCCESSIVE SAMPLING DATES AT THREE SITES

4.3.2.2. Distribution in space

The difference in numbers of each pest in samples taken from two locations: in the row centred on seedlings and between the rows, was analysed by Analysis of Variance or Students 't' test. Results showed that generally all the pests studied could be found in greater numbers in the row around seedlings but differences were not always significant. Since different sizes of sample unit were used, comparisons of numbers found are not valid between sites.

Fig. 7 shows the effect of sample location on the distribution of millipedes. At Shouldham in 1972 the early sample in May, taken with a 2.3 in. diam. auger each enclosing one complete seedling, showed numbers in the two sampling locations to be non-significant whereas the later sample in June, using bulked cores each centred on and including 10 seedlings, indicated a highly significant increase in millipede numbers in the row than midway between rows. At Marham and Kettering sites samples were also taken on two dates and the same sized sampling unit was used (8 cores, each centred on a seedling). In addition to an increase in the numbers of millipedes in the row at the latter sampling date, differences between sampling location were significant on both dates.

Fig. 8 shows the effect of sample location on the distribution of pygmy beetle. All samples showed the difference between 'in row' and 'between row' numbers to be highly significant ($P < 0.1\%$). At March, Boston and Welney sites the sample unit was a single core, each 2.3 in. diam. and enclosing a complete seedling. At Welney a period of 5 days separated first and second sampling-times; the numbers of beetles around the seedlings increased whereas numbers

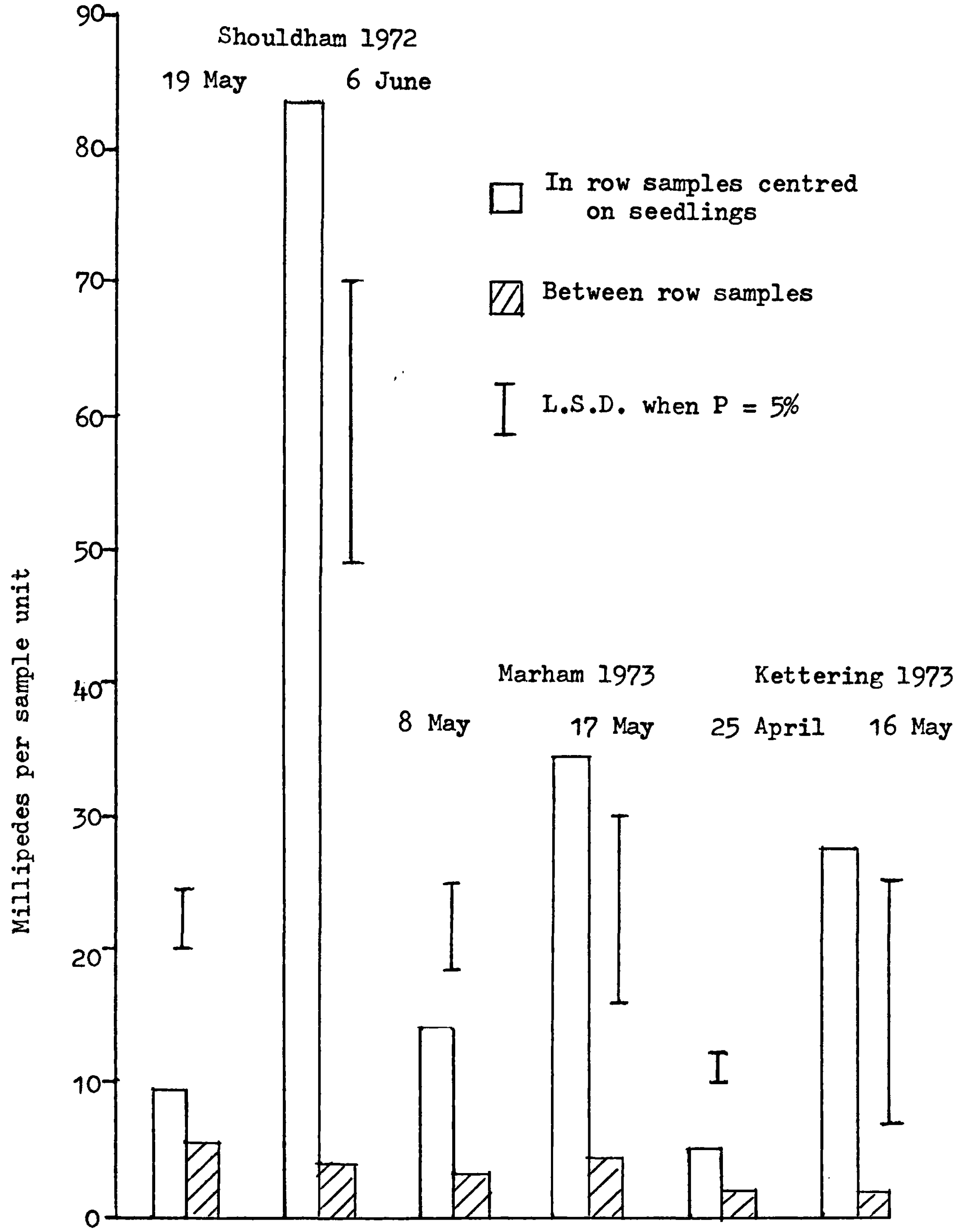


FIG. 7. EFFECT OF SAMPLE LOCATION ON THE DISTRIBUTION OF MILLIPEDES IN THE CROP.

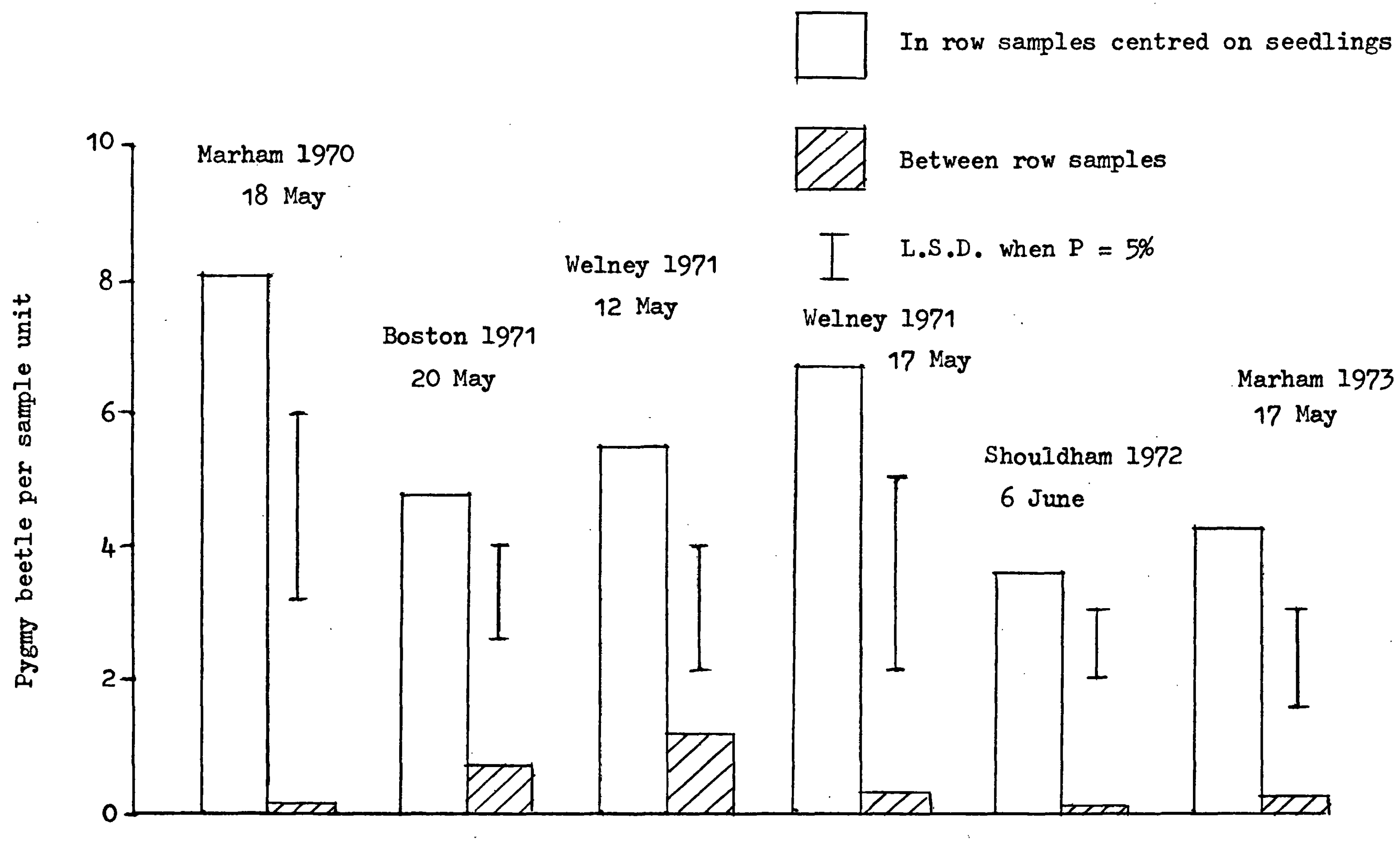


FIG.8. EFFECT OF SAMPLE LOCATION ON THE DISTRIBUTION OF PYGMY BEETLE IN THE CROP.

found midway between the rows decreased. At Marham in 1973, where beet was grown for the second consecutive year, the number of beetles found in the crop were no more than at Shouldham in 1972 where the beet was grown in the normal rotation.

Fig. 9 shows the effect of sample location on the distribution of Collembola. At Welney 1971, Broom's Barn 1972 and Shouldham 1972 the histograms refer to Onychiuridae which includes Onychiurus spp. and Tullbergia spp. mainly. Onychiurus spp. were separated from the remaining Collembola at the other sites. The histograms of Kettering, Littleport and the April Broom's Barn samples refer to the number of Collembola per 2.3 x 3.9 in. soil core and hence numbers are comparable between sites. Of the five sites the numbers in the row at only two, Stalham and Broom's Barn, were significantly more than in samples taken midway between the rows - these were the only sites where Onychiurus spp. were thought to be causing damage to the seedlings. Most Collembola were found at Broom's Barn in samples taken on 26 March shortly after sowing and before the seedlings had emerged. At Stalham the field was sampled when the seedlings were in the late-cotyledon stage but many had failed to emerge leaving large gaps in the rows. At Kettering and Littleport there were significant differences between sample location, being highly significant ($P < 0.1\%$) at the former site; here, millipedes were also found in the seedling root zone but in only low numbers at this time (average of 5 per sample unit of the same size). At Littleport the Onychiurus spp. occurred together with Symphyla (Scutigerella immaculata) but at the Broom's Barn site no other pest was present even in the late (17th April) samples.

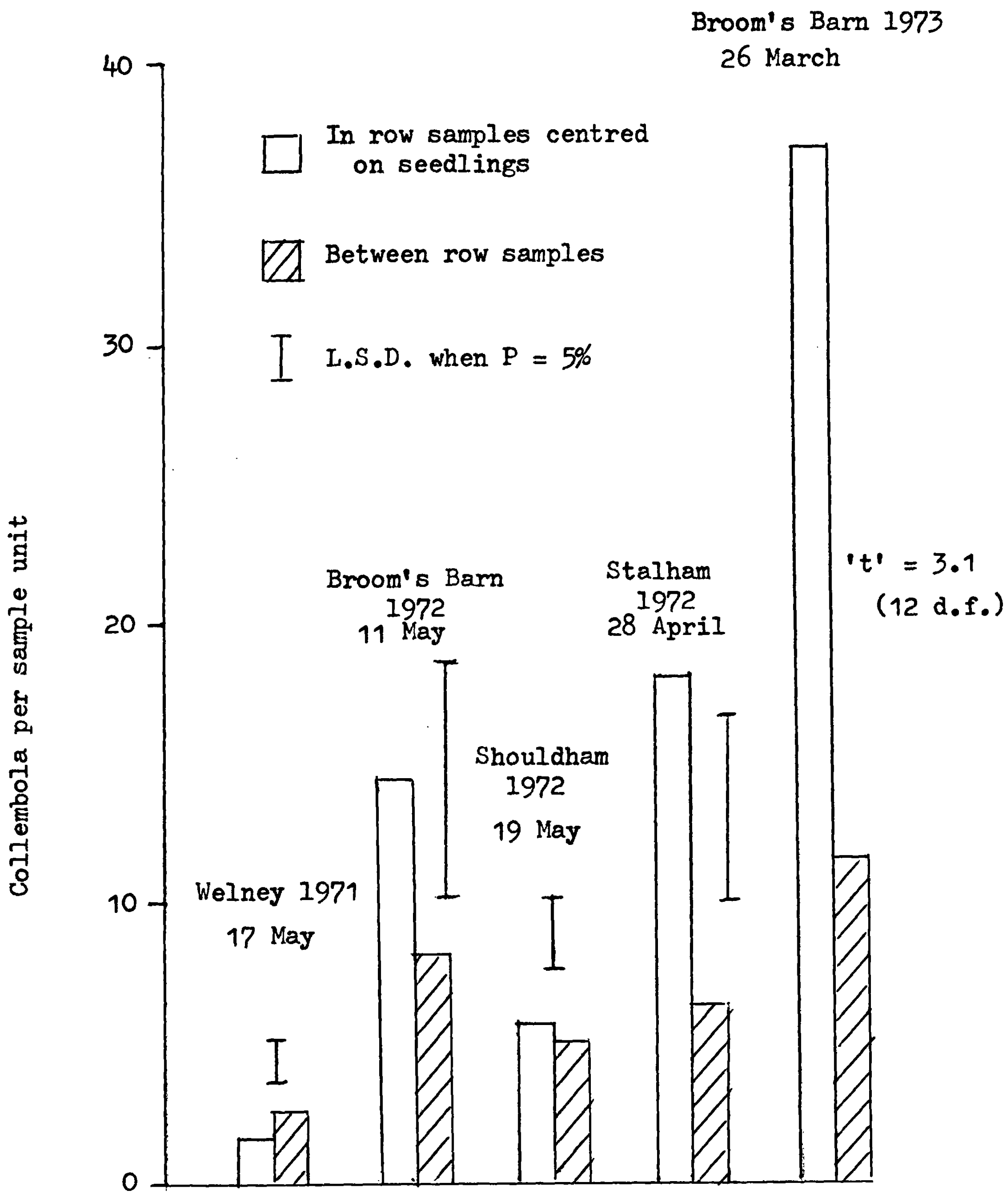


FIG.9. <u>EFFECT OF SAMPLE LOCATION ON THE DISTRIBUTION OF COLLEMBOLA IN THE CROP.</u> Continued on P. 68

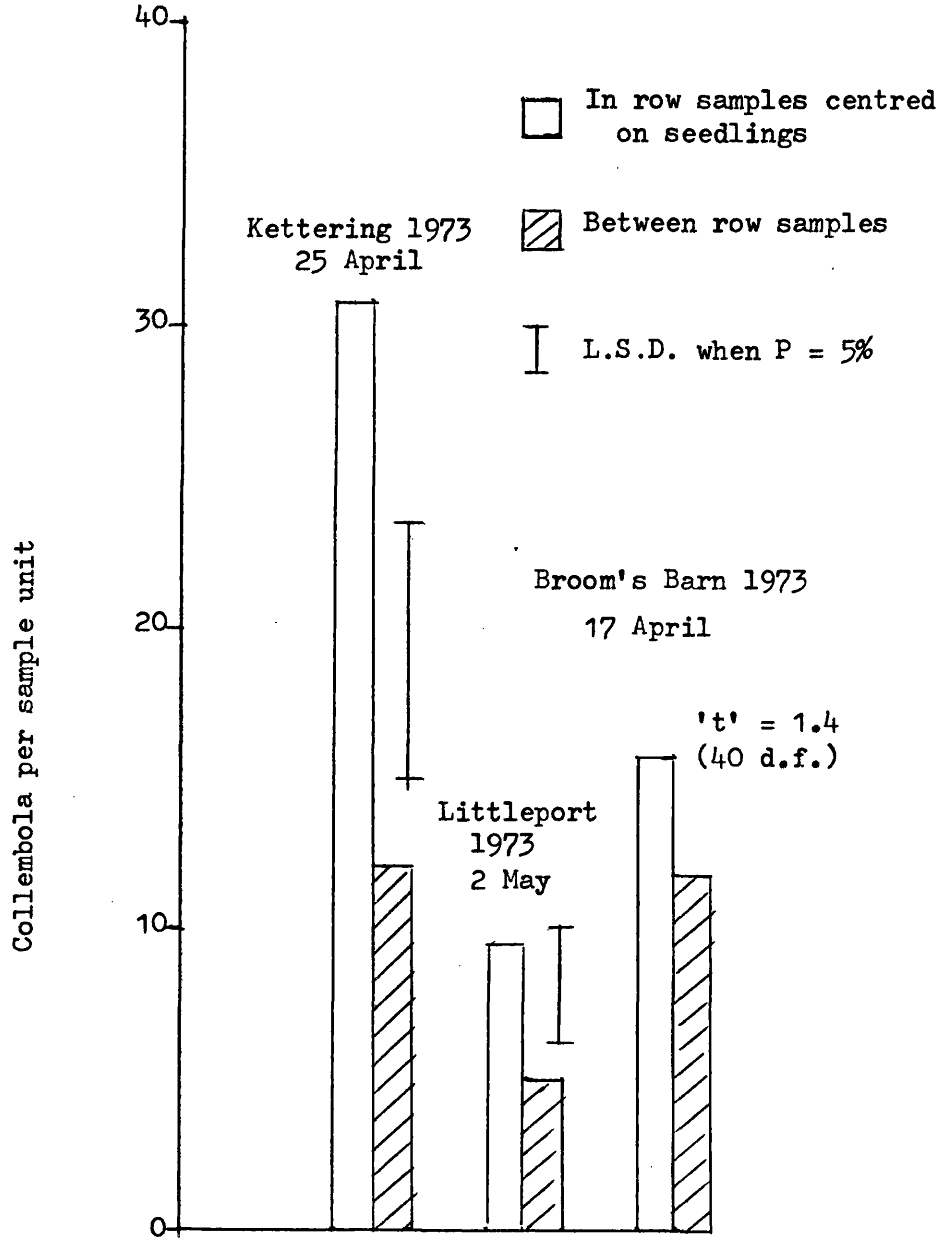


FIG. 9. (CONTINUED) EFFECT OF SAMPLING LOCATION ON THE DISTRIBUTION OF COLLEMBOLA IN THE CROP.

4.3.2.3. Dispersion

The dispersion of a population (the pattern of the animals' distribution in space) is an important ecological concept since it affects both the sampling programme and statistical treatment of the data. The data collected from taking individual soil cores both in bare soil and centred on a seedling was analysed from many sites to find the variance and mean for each sample for each of three groups of pests: millipedes, pygmy beetle and Collembola. The relationship between these two parameters describes their pattern of distribution in the soil; if variance is equal to the mean then their distribution is random and conforms to the Poisson series; if the variance is less than the mean they are more regularly distributed, but if variance is more than the mean a 'contagious' or aggregated distribution is implied. The extent to which the distribution of millipedes, pygmy beetle and Collembola satisfied the Poisson model was tested by χ^2 and a Dispersion Index obtained, a statistic given by Bliss & Fisher (1953) and Healy (1962). This was, in effect, a test for aggregation:

$$\chi^2 = \frac{s^2(N-1)}{\bar{x}}$$

where s^2 = variance; N = number of sample units; $\bar{x}$ = the sample mean.

The test was applied to both 'in row' and 'between row' samples of each pest. A non-significant value of χ^2 implies that the distribution can be considered as random. The degree of aggregation was also calculated using the Lexis Index of Aggregation, λ :

$$\lambda = \frac{s}{\bar{x}}$$

Tables 11 - 16 give the population parameters for each of the pests, Dispersion Indices and the departure from the Poisson or randomly distributed population, the significance of which was denoted by asterisks.

Millipedes were always highly aggregated both in bare soil (Table 11) and around seedling roots (Table 12). Stadium II millipedes aggregated markedly more than adult stages at Shouldham, the index value decreased in consecutive samples from 3.8 on 6 April to 3.1 on 20 April, to 2.1 on 25 May; conversely adult stadia (VII and above) became more aggregated.

Pygmy beetle were found to be aggregated in bare soil (Table 13) in two of the five samples and in three of the four 'in row' samples which included the root zone of included seedlings (Table 14).

The aggregation indices for Collembola ranged from a non-significant 1.1 to a highly significant 4.0 in bare soil and the overall mean in bare soil was 2.3 compared with the corresponding value for millipedes of 2.9 (Table 15). A highest value of 5.4 was achieved in the root zone of seedlings (Table 16).

The individual sample means were plotted against the corresponding Lexis Index of Aggregation for each pest and included values obtained from bare soil and root zone aggregates. For each pest the Aggregation Index increased with an increase in numbers of pests per core. A Regression Analysis of the data for each pest fitted straight lines to the graphs. The line for millipedes was less steep than for pygmy beetle (Fig.10) or Collembola (Fig.11) indicating that they are more aggregated at any given population density; Collembola were, comparatively, the least aggregated.

TABLE 11. MILLIPEDE DISPERSION IN BARE SOIL

Site	Sampling date	Sample mean $\bar{x}$	No. of samples N	Dispersion Index χ^2	Lexis Index λ
SHOULDHAM					
a Total Millipedes	6.4.72	8.6	30	393.5***	3.7
b Stadium II	"	7.6	30	408.7***	3.8
c Adults only	"	1.0	30	37.7	1.1
SHOULDHAM					
a Total Millipedes	20.4.72	8.9	48	503.3***	3.3
b Stadium II	"	6.6	48	460.0***	3.1
c Adults only	"	2.0	48	105.8***	1.5
SHOULDHAM					
a Total Millipedes	25.5.72	4.2	30	149.8***	2.3
b Stadium II	"	1.9	30	132.8***	2.1
c Adults only	"	1.5	30	100.5***	1.9
SHOULDHAM					
Total Millipedes	20.6.72	6.7	30	435.9***	3.9
MARHAM					
a Total Millipedes	12.4.73	2.1	24	37.2*	1.3
b Stadium II	"	2.3	24	58.0***	1.6
c Adults only	"	1.1	24	85.7***	1.9

TABLE 12. MILLIPEDE DISPERSION IN THE ROW AROUND SEEDLING ROOTS

Site	Sampling date	$\bar{x}$	N	χ^2	λ
SHOULDHAM					
a Total Millipedes	19.5.72	7.3	48	511.8***	3.3
b Stadium II	"	2.6	48	218.7***	2.2
c Adults only	"	4.3	48	450.3***	3.1

*, ***, denotes probability at P <5%, 0.1% that distribution is not of the Poisson type.

TABLE 13. PYGMY BEETLE DISPERSION IN BARE SOIL

Site	Sampling date	Sample mean $\bar{x}$	No. of samples N	Dispersion Index χ^2	Lexis Index λ
WELNEY	a 19.4.71	1.2	32	74.9***	1.6
	b 17.5.71	1.1	24		1.0
BOSTON	20.5.71	0.7	48	241.7***	2.4
MAGDALEN	5.5.72	0.5	24	27.6	1.1
BENWICK	27.3.72	1.2	24	23.0	1.0

TABLE 14. PYGMY BEETLE DISPERSION IN THE ROW AROUND SEEDLINGS

Site	Sampling date	Sample mean $\bar{x}$	No. of samples N	Dispersion Index χ^2	Lexis Index λ
WELNEY	a 17.5.71	6.7	24	172.0***	2.7
	b 17.5.71	5.5	24	84.5***	1.9
BOSTON	20.5.71	4.8	48	134.1***	1.7
MAGDALEN	5.5.72	1.3	24	30.1*	1.2

*, ***, denotes probability at P <5%, 0.1% that distribution is not of the Poisson type.

TABLE 15. DISPERSION OF COLLEMBOLA (ONYCHIURIDAE AND ONYCHIURUS SP.) IN BARE SOIL

Site	Sampling date	Sample mean $\bar{x}$	No. of samples N	Dispersion Index χ^2	Lexis Index λ
WELNEY	19.4.71	5.5	32	146.0***	2.2
WELNEY	a 17.5.71	1.7	24	44.6**	1.4
WELNEY	b 17.5.71	2.3	24	59.0***	1.6
BOSTON	20.4.71	5.2	32	506.1***	4.0
BOTTISHAM	13.4.71	3.6	25	61.3***	1.6
BOTTISHAM	4.6.71	1.3	16	25.4*	1.3
BROOM'S BARN	11.5.72	7.8	24	224.4***	3.1
BENWICK	27.3.72	10.4	24	118.5***	2.3
MAGDALEN	30.3.72	3.8	24	81.7***	1.9
MAGDALEN	5.5.72	0.9	24	28.1	1.1
BROOM'S BARN	10.4.73	3.5	24	57.8***	1.6
LITTLEPORT	30.3.73	7.0	24	111.1***	2.2
KETTERING	17.3.73	16.0	24	564.9***	5.0

TABLE 16. COLLEMBOLA DISPERSION IN THE ROW AROUND SEEDLINGS

Site	Sampling date	Sample mean $\bar{x}$	No. of samples N	Dispersion Index χ^2	Lexis Index λ
WELNEY	a 17.5.71	2.9	24	178.4***	2.8
WELNEY	b 17.5.71	1.5	24	76.7***	1.8
BOTTISHAM	4.6.71	1.9	16	18.2	1.1
BROOM'S BARN	11.5.72	14.1	24	679.4***	5.4
MAGDALEN	5.5.72	1.2	24	30.7	1.2

*, **, ***, denotes probability at P <5%, 1%, 0.1% that distribution is not of the Poisson type.

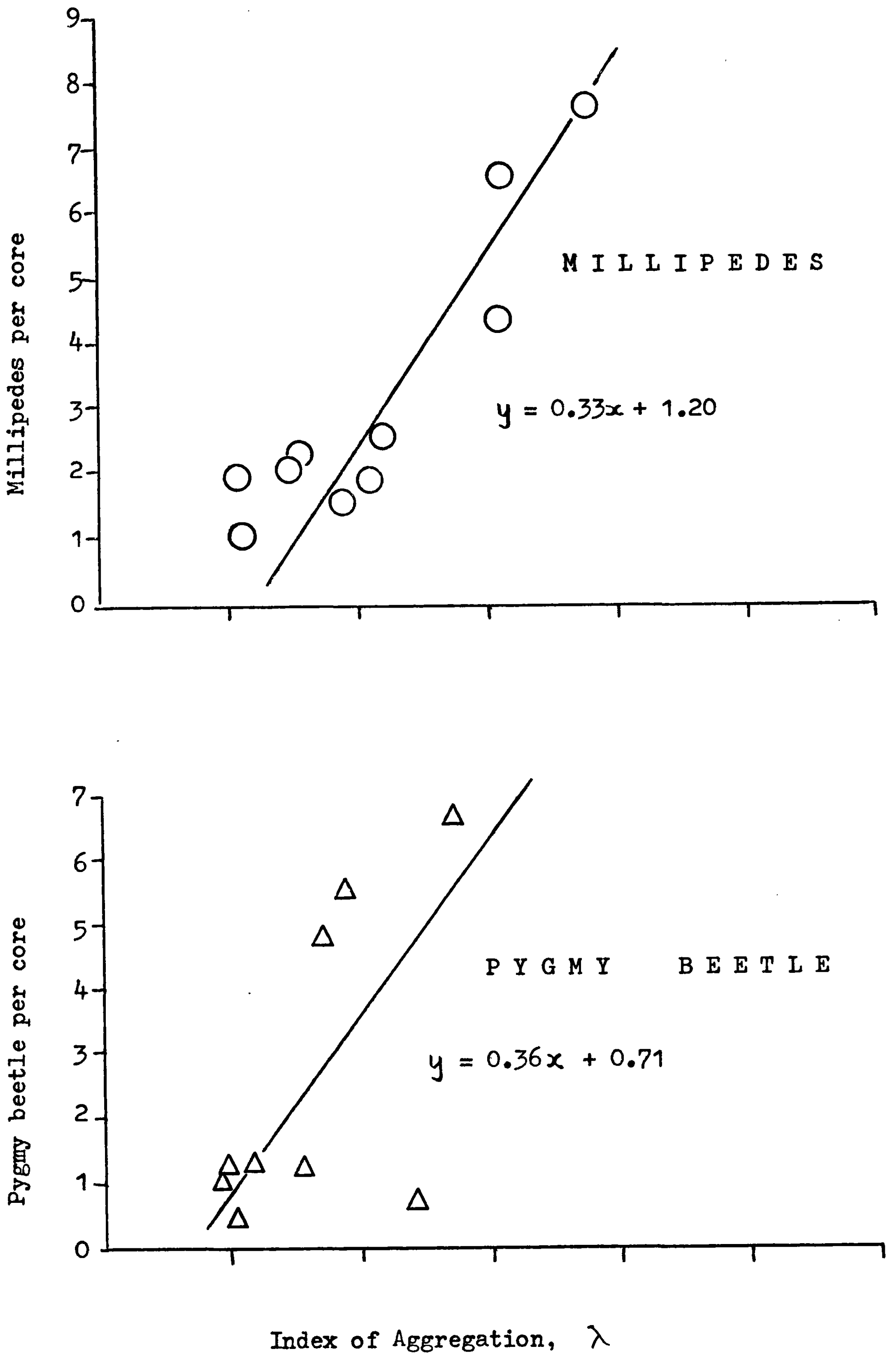


FIG. 10. THE RELATIONSHIP BETWEEN PEST DENSITY AND THE DEGREE OF AGGREGATION OF MILLIPEDES AND PYGMY BEETLE IN THE SOIL.

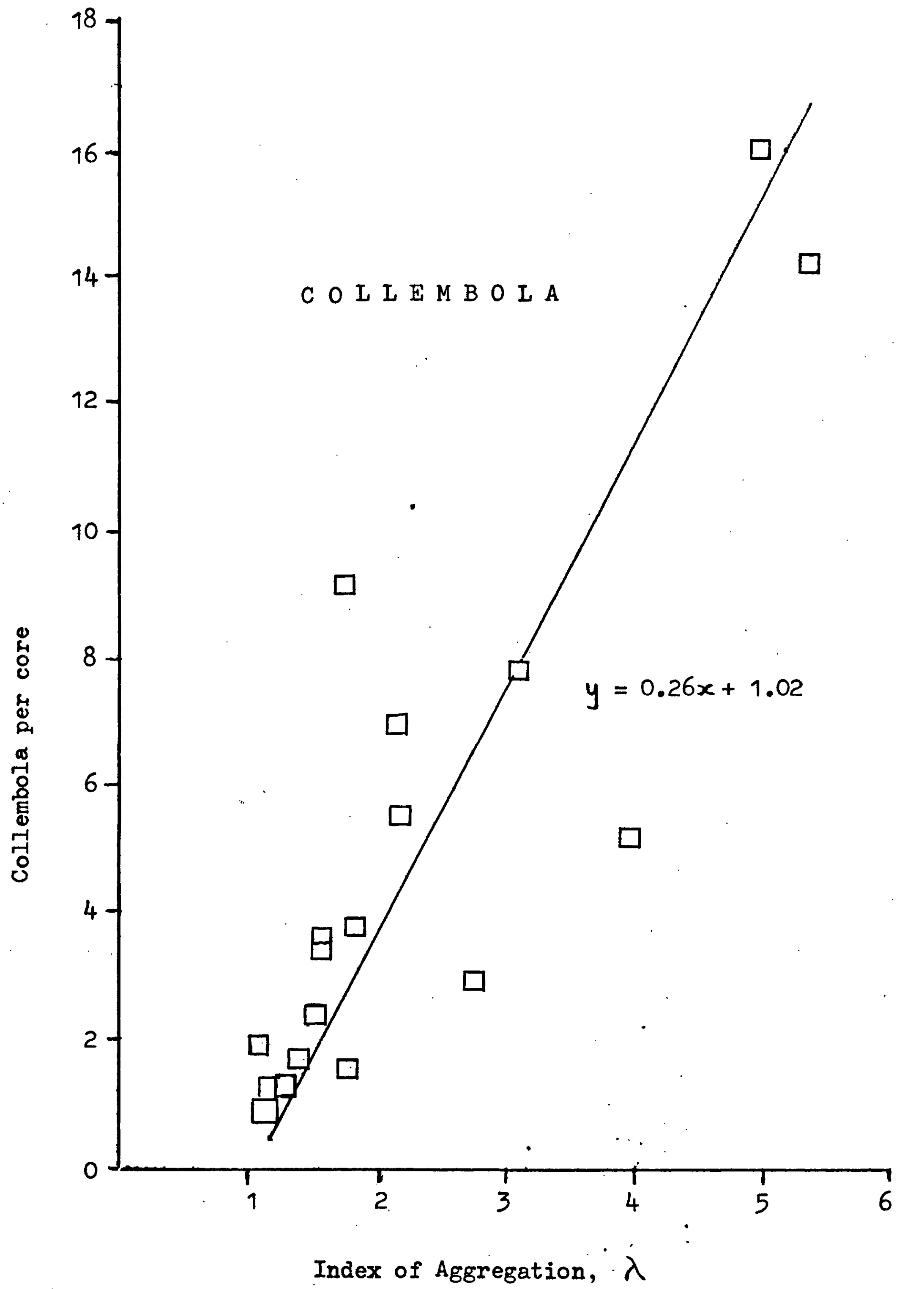


FIG. 11. THE RELATIONSHIP BETWEEN THE DENSITY AND THE DEGREE OF AGGREGATION OF COLLEMBOLA IN THE SOIL.

4.3.3. Effect of seedling spacing on pest distribution in the rows.

4.3.3.1. Statistical aspects.

Although a herbicide treatment was included in the 1971, 1972 and some 1973 experiments it was evident that weed growth on control plots was so sparse as to render them indistinguishable from the sprayed plots. In the analysis of the data from experiments at Marham and Broom's Barn in 1973 the plots both with and without herbicide were considered as one treatment thus increasing the effective replication in the treatment of the data. All data was analysed after a Log (n+1) transformation using Analysis of Variance but the spacing and herbicide effects were separated in the analysis of the remaining experiments and presented separately in the results.

Soil sampling from 1971 to 1972 illustrated the variability in numbers of pests in the root zone but there was little opportunity to calculate the number of cores needed. At Shouldham in 1972 the first sample taken on the seed spacing experiment (19 May sample) was used to obtain an estimate of the number of samples required by calculating the mean and variance of all the 'in row' sample cores. The mean of 9.25 and variance of 56.08 emphasized the variability within the sample. It can be seen that any treatment effect that may have been present was masked by the variation between blocks (Table 17) and the 'contagious' type or non-random distribution of the millipedes.

TABLE 17. DATA FROM ANALYSIS OF VARIANCE OF SEEDLING SPACING EFFECTS ON MILLIPEDE DISTRIBUTION

Site		Shouldham		Marham			Kettering			Bottisham
Date		19.5.72	6.6.72	8.5.73	17.5.73	6.6.73	25.4.73	14.5.73	30.5.73	14.5.74
Variance Ratio, F.	Blocks	2.5	2.0	0.2	9.6**	1.6	0.2	0.1	1.0	23.2**
	Spacings	0.3	8.8**	1.9	3.2	0.8	0.1	1.8	7.6*	0.6
Coefficient of Variation (%)		43.0	16.1	29.8	19.9	46.0	35.2	29.5	30.0	56.0

*, **, denotes differences significant at P <5%, 1% respectively.

An estimate of the number of sample units N, needed to give an accuracy of 10% was calculated from this preliminary sample at Shouldham by adoption of the relationship:

$$N = \frac{ts}{D\bar{x}}^{2}$$

where: s = standard deviation

D = required level of accuracy (normally 0.1)

't'= quantity depending on number of samples and is obtained from tables; approximates to 2 at 5% level for more than 10 samples. The mean and variance values were substituted and the value of N was calculated to be 161. In order to reduce both the time taken to sample and process the cores it was decided to bulk the cores into units of 10; this would not affect the numbers of creatures recovered but would reduce the number of degrees of freedom in the analysis.

The subsequent soil sample with 160 cores per treatment showed that the differences between treatments was highly significant since the Variance ratio, F, was 8.8 with 2 and 15 degrees of freedom (Table 17). The block component, though still high, was not significant due to the increased size of the sample. As a result of this exercise a rough estimate was obtained of the sample size to which to aim; future sample cores were bulked into more convenient units of 8. A constraint on the sample size was provided by the necessity of sampling on two or three successive dates on each plot. The effect would be to seriously affect the number of seedlings available and it was not possible to increase the plot size without reducing the number of replicates. For these reasons the number of cores taken was reduced to usually 64 or 72 per treatment. Variance ratios of both block and spacing components were calculated to assess the effectiveness of the replication and contribution of the treatments to the variation within the data.

4.3.3.2 Millipedes

Blaniulid millipedes were found only at four sites in numbers sufficient for sampling seedling spacing experiments. The first sample at Shouldham, taken on 19 May 1972, failed to demonstrate either a significant aggregation in the rows or any difference between seedling spacings, viz: 11.0, 8.9 and 7.9 millipedes per core for seedlings spaced at 1.5, 4.5 and 9.0 in. The later sample using the improved sampling technique showed that the numbers around the seedlings was inversely proportioned to seedling spacing with significantly more millipedes around seedlings spaced at 9.0 and 4.5 in. than at 1.5 in. (Fig.12).

At Marham the number of millipedes in the root zone of seedlings taken from all three spacings was greater in the second sample taken on 17 May with significantly more millipedes around seedlings spaced at 4.5 in. than at 1.5 in. but no difference between numbers around seedlings at the two wider spacings (Fig. 13). The difference between blocks was highly significant ($P < 0.01$) at this site (Table 17), indicating that the millipede distribution in the field was extremely variable over the trial area. The number of millipedes recovered from the last sample taken on 6 June at the time of singling, when the top soil was becoming very dry, was fewer than the previous

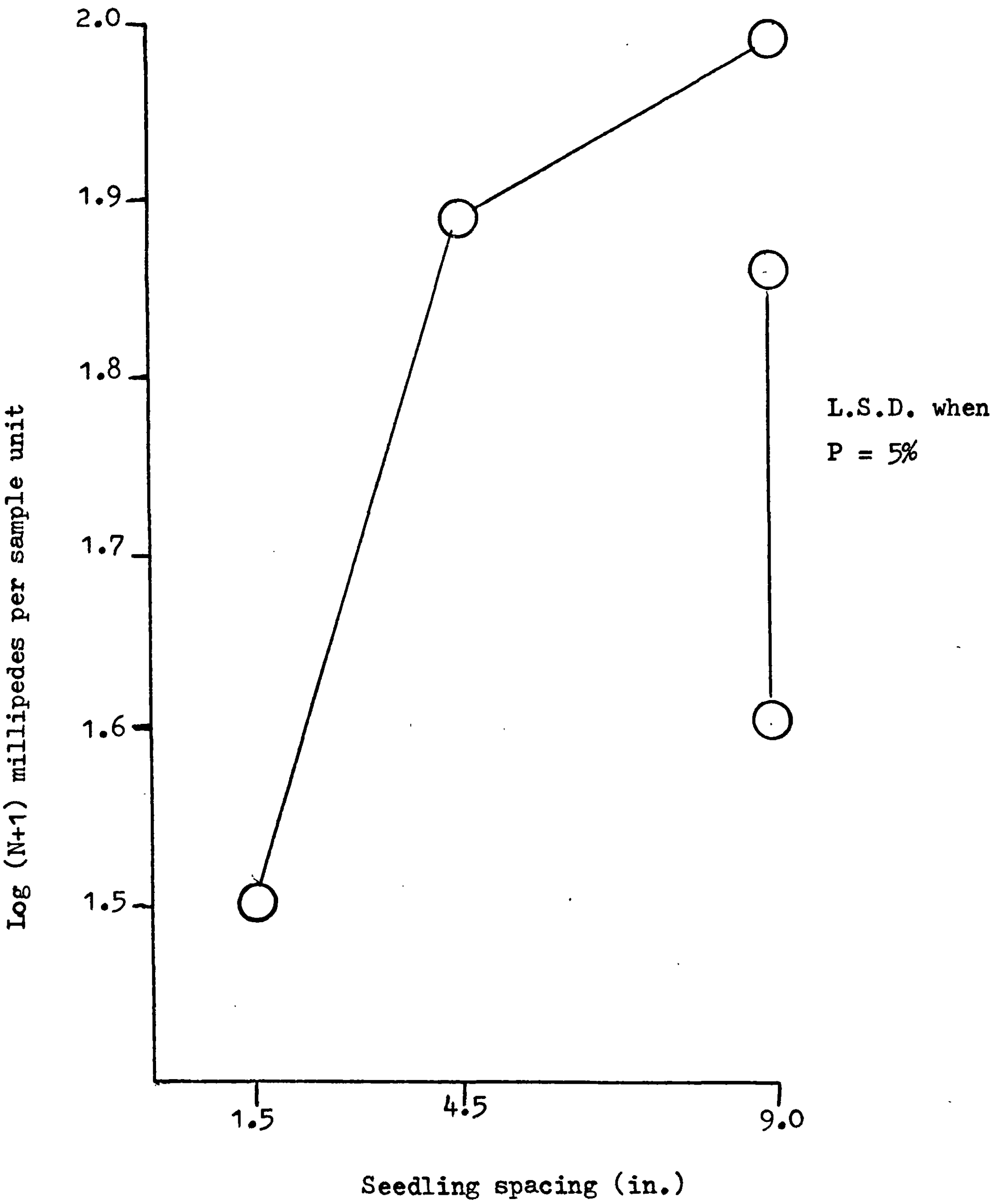


FIG. 12. EFFECT OF SEEDLING SPACING ON NUMBERS OF MILLIPEDES IN THE ROOT ZONE (SHOULDHAM, 6 JUNE, 1972).

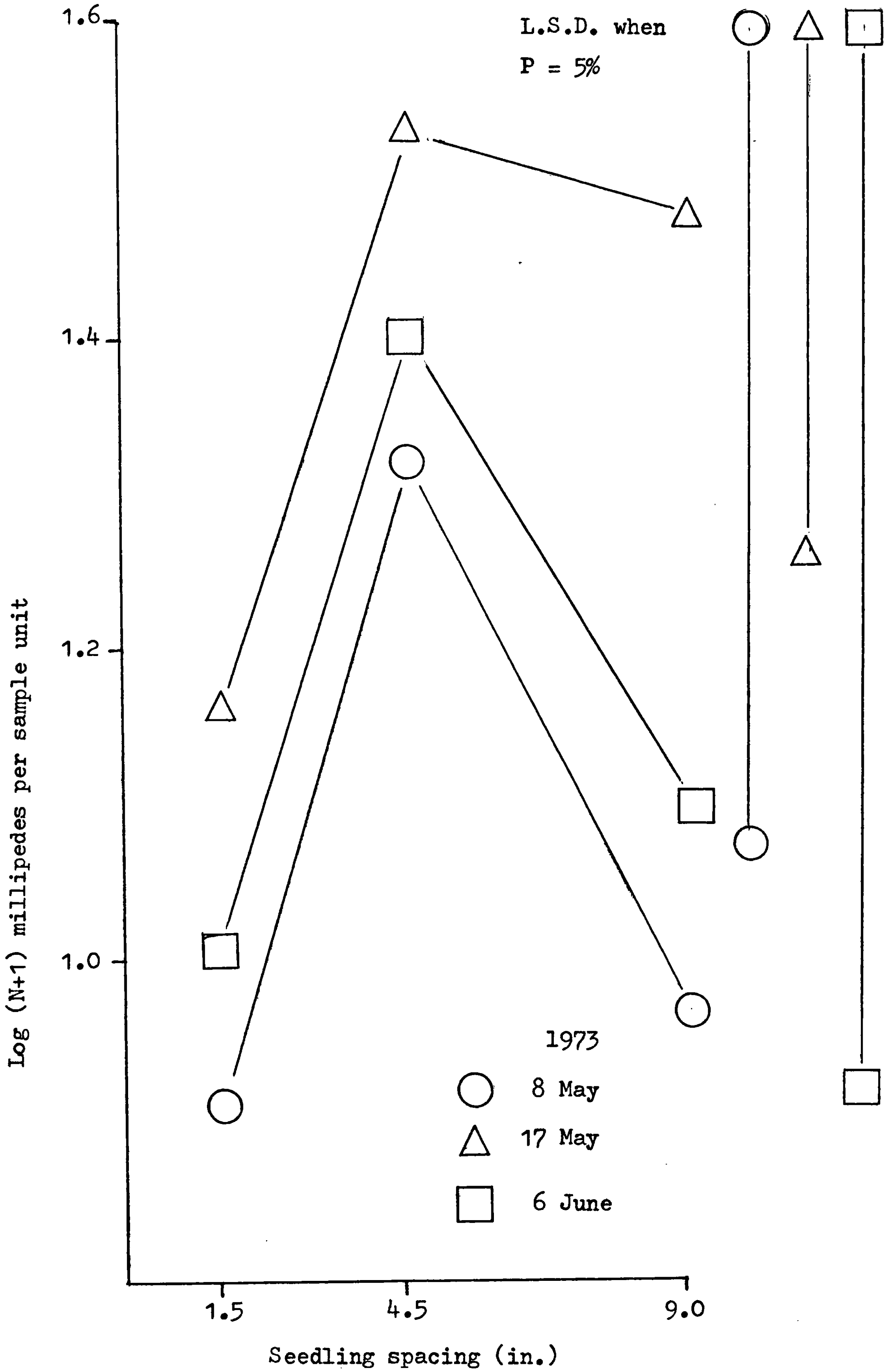


FIG. 13. EFFECT OF SEEDLING SPACING ON NUMBERS OF MILLIPEDES IN THE ROOT ZONE ON THREE SAMPLING DATES (MARHAM, 1973).

sample for all spacings with the largest decrease in numbers from the widest spaced seedlings. There were no significant differences in millipede numbers at the latest sampling date.

At Kettering numbers of millipedes around seedlings at all spacings similarly increased between first and second sampling dates, 25 April to 14 May respectively, (Fig.14). In these samples there was no significant difference between numbers of millipedes around seedlings at any of the spacings. The last sample taken on 30 May at the time of singling showed a fall in numbers on all treatments, as at Marham, particularly in numbers around the closest spaced seedlings; this was sufficient to establish a significant difference between numbers of millipedes around seedlings at 1.5 in. and 9.0 in.

At Bottisham in 1974 the block variation was significant ($P < 0.1$) throughout the length of the experiment (Table 17) and many more millipedes were found at one end of the experiment, where damage to the sugar-beet seedlings had occurred in 1970. Although single row plots were used to increase replication and make some allowance for small patches of infestation the difference in numbers between treatments was not significant, viz: 3.6, 4.4 and 7.8 millipedes per sample unit for seedling spacings of 1.5, 4.5 and 9.0 in. respectively.

4.3.3.3 Pygmy beetle.

Fig. 15 shows the effect of seedling spacing on the numbers of pygmy beetle in the root zone at Shouldham in 1972 and at Kettering and Littleport in 1973. There was no difference in numbers of beetles on the three spacings in the Shouldham samples

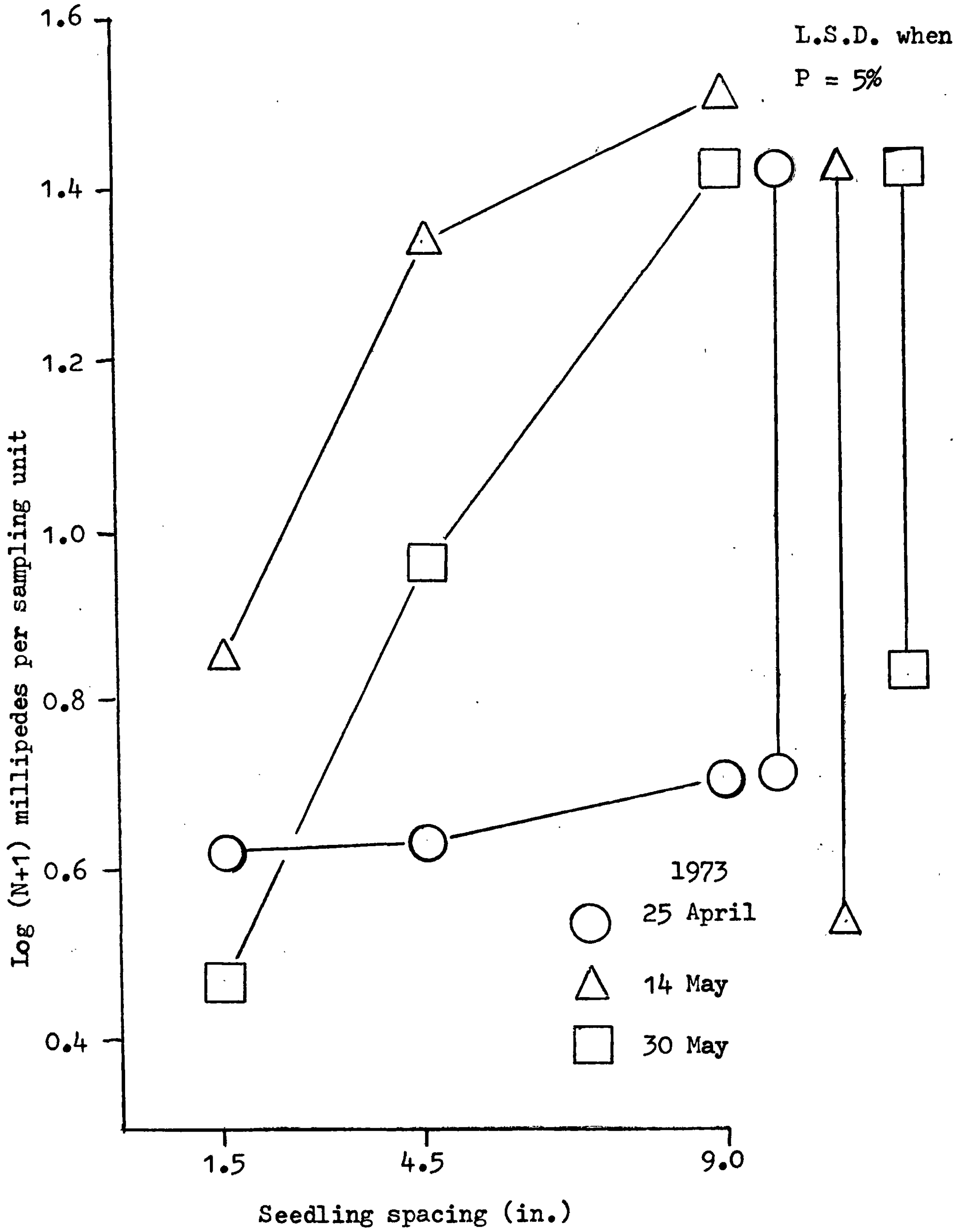


FIG. 14. EFFECT OF SEEDLING SPACING ON NUMBERS OF MILLIPEDES IN THE ROOT ZONE ON THREE SAMPLING DATES (KETTERING, 1973).

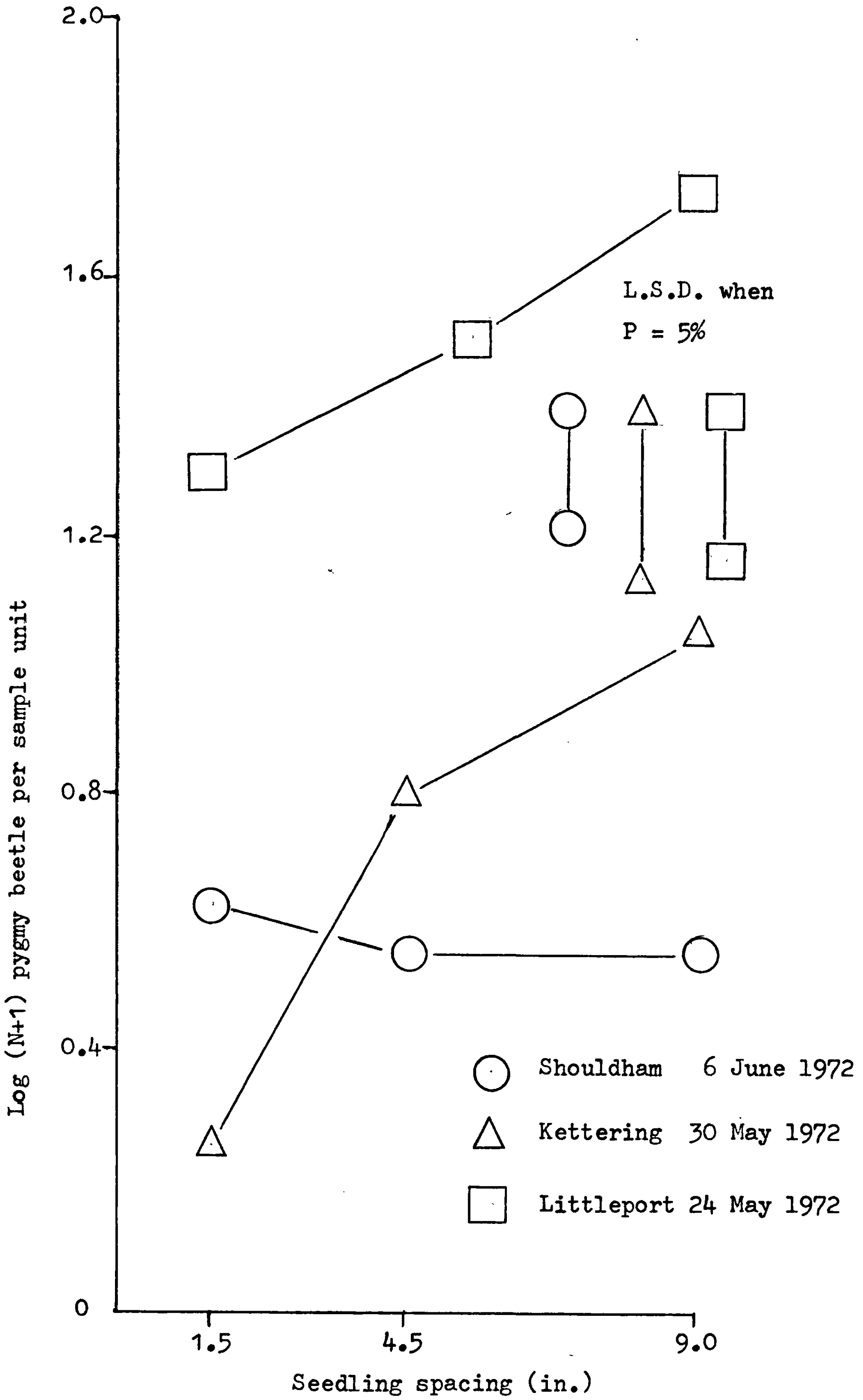


FIG. 15. EFFECT OF SEEDLING SPACING ON NUMBERS OF PYGMY BEETLE IN THE ROOT ZONE AT THREE SITES.

but there were differences at the other two sites. Differences were significant between 1.5 and 4.5 in. spacings and 1.5 in. and 9 in. spacings at Kettering and between 4.5 in. and 9 in. and 1.5 in. and 9 in. spacings at Littleport. At Marham the results of sampling on two consecutive dates are shown in Fig.16. Whereas the mean number of beetles found around seedlings at 1.5 in. was almost exactly the same on the two sampling dates, numbers around seedlings at 4.5 in. and 9.0 in. had increased but differences were not significant. At Shouldham site the block component was significant (Table 18) indicating a heterogenous distribution over the experimental area.

4.3.3.4 Collembola.

Fig. 17 shows the effect of seedling spacing on numbers of Collembola in the root zone on three different fields at Broom's Barn from 1971 to 1973. In 1971 and 1972 the Collembola recorded were <u>Onychiuridae</u> but in the 1973 sample <u>Onychiurus</u> was recorded specifically. In the 1971 sample a seed spacing of 3 in. instead of 4.5 in. was sampled; there were no significant differences in numbers of Onychiuridae around seedlings at the different spacings. In 1972 significantly more Onychiuridae were found around seedlings spaced at 1.5 in. than 4.5 in. In 1973 the numbers of <u>Onychiurus</u> around seedlings at 1.5 in, 4.5 in. and 9 in. was similar with no significant differences. Similarly in samples taken at Kettering, Littleport and Stalham sites (Fig.18) no significant differences were detected. At all sites the block variation was significant (Table 19).

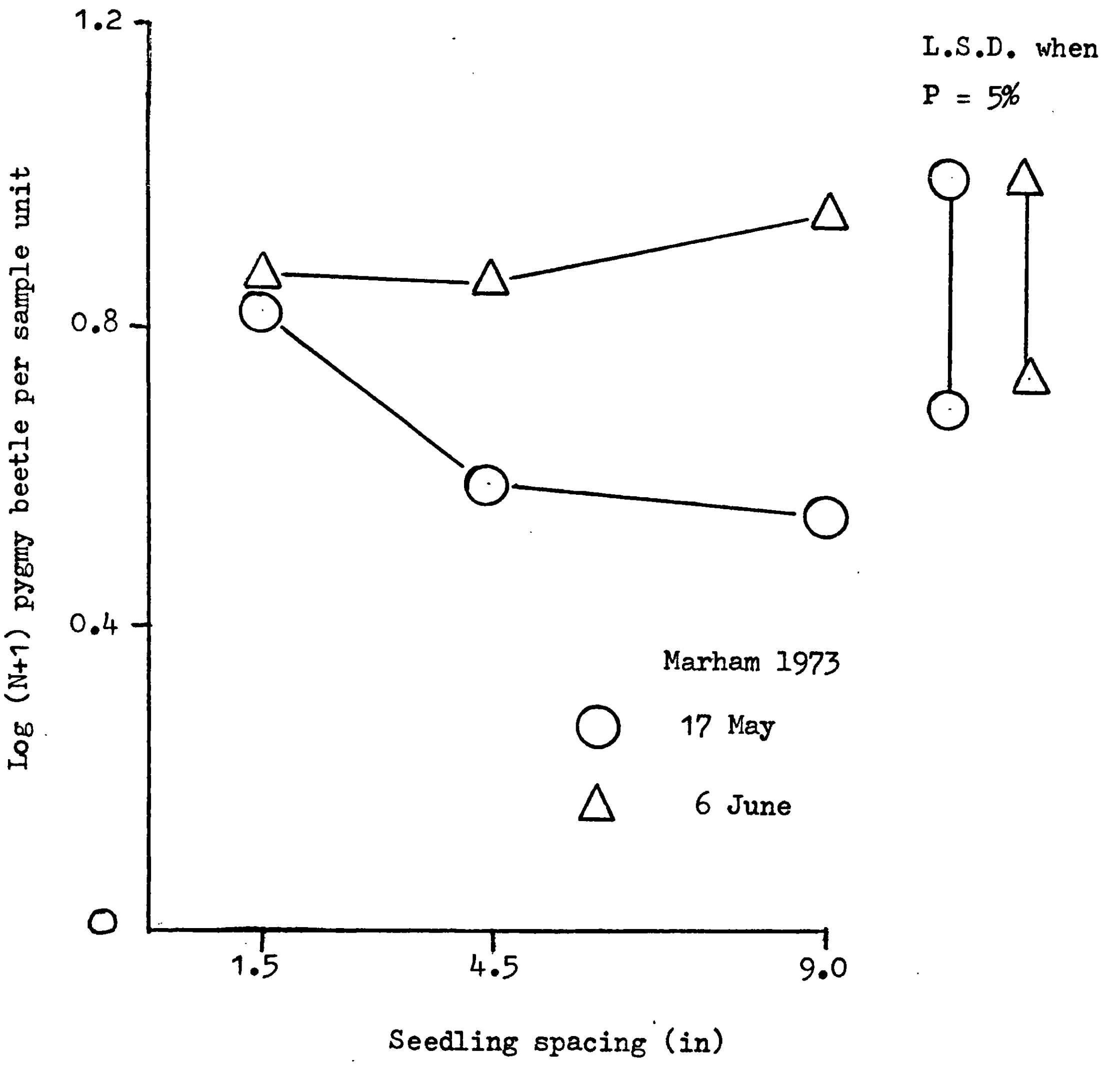


FIG. 16. EFFECT OF SEEDLING SPACING ON NUMBERS OF PYGMY BEETLE IN THE ROOT ZONE ON TWO SAMPLING DATES.

TABLE 18. DATA FROM ANALYSIS OF VARIANCE OF SEEDLING SPACING EFFECTS ON PYGMY BEETLE DISTRIBUTION

Site		Shouldham	Kettering	Littleport		Marham	
Date		6.6.72	30.5.73	2.5.73	24.5.73	17.5.73	6.6.73
Variance Ratio, F	Blocks	5.9*	1.5		3.8	2.7	2.8
	Spacings	0.6	28.5**		11.5**	2.2	0.3
Coefficient of variation (%)		43.0	31.6		10.8	37.9	24.5

*, **, denotes differences significant at P <5%, 1% respectively

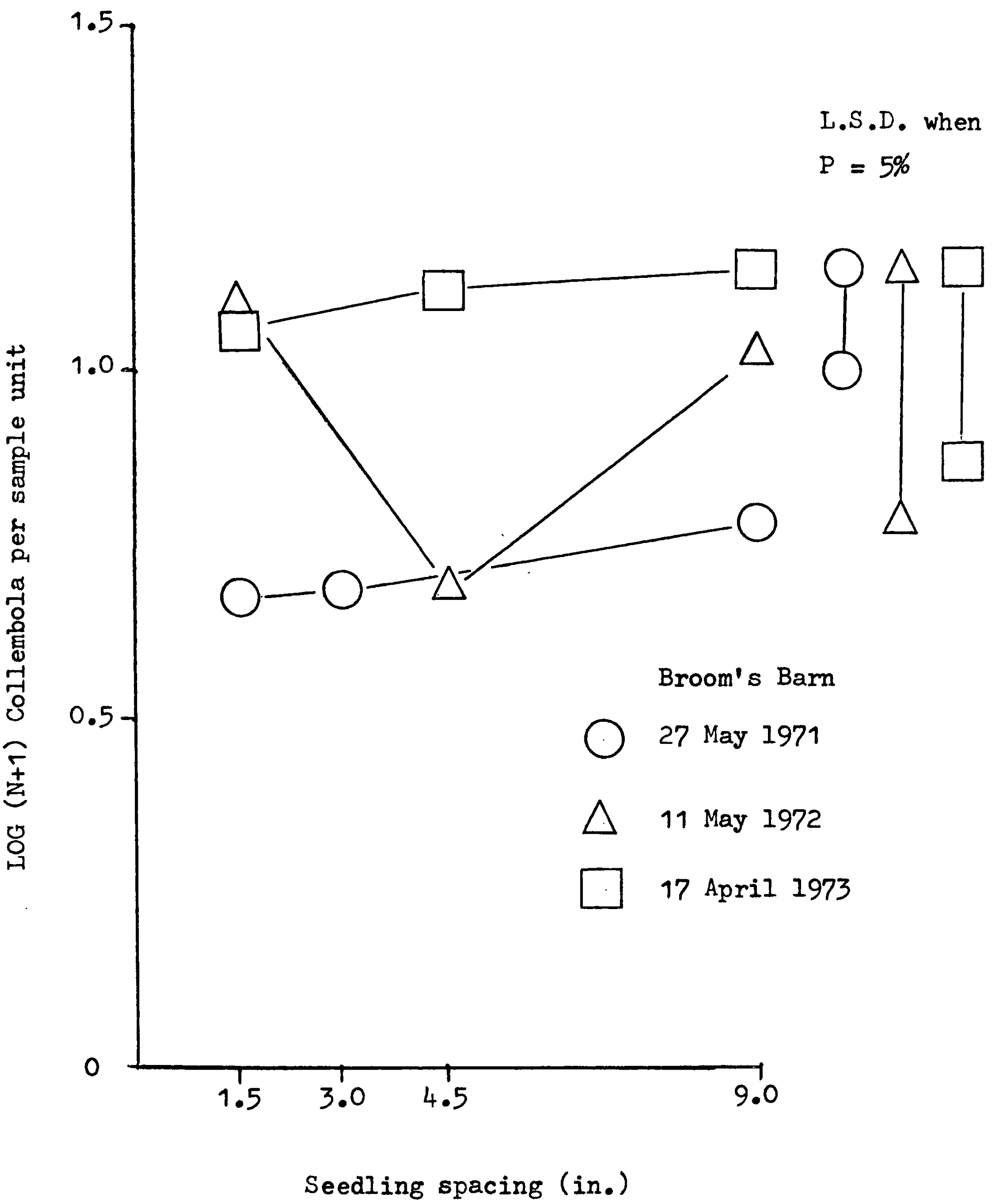


FIG. 17. EFFECT OF SEEDLING SPACING ON NUMBERS OF COLLEMBOLA IN THE ROOT ZONE ON THREE FIELDS AT BROOM'S BARN.

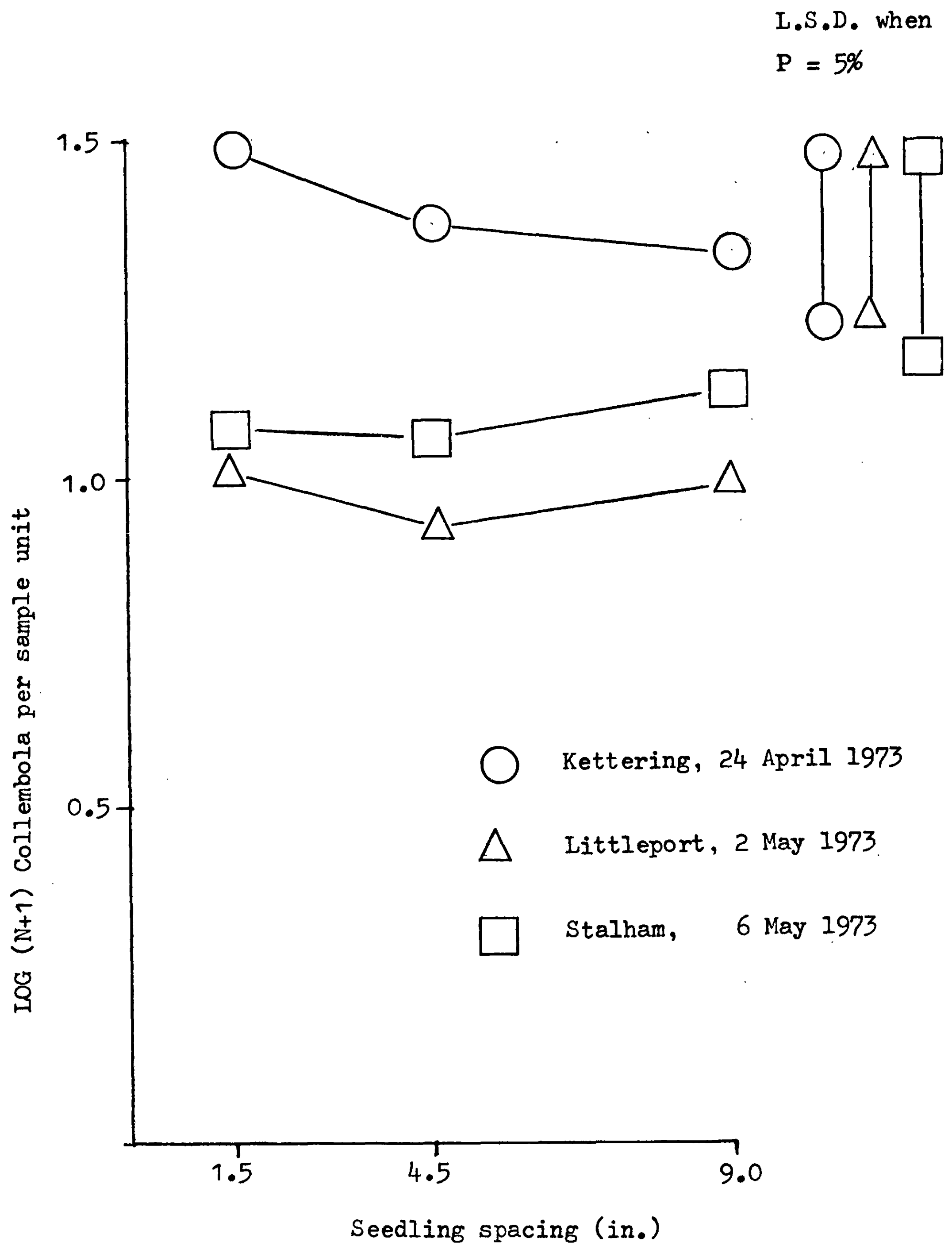


FIG. 18. EFFECT OF SEEDLING SPACING ON NUMBERS OF COLLEMBOLA IN THE ROOT ZONE AT THREE SITES.

TABLE 19. DATA FROM ANALYSIS OF VARIANCE OF SEEDLING SPACING EFFECTS ON COLLEMBOLA DISTRIBUTION.

Site		Broom's Barn		Kettering	Littleport	Stalham
Date		11.5.72	17.4.73	25.4.73	2.5.73	6.5.74
Variance Ratio, F.	Blocks	5.7*	4.9*	15.2**	6.0	11.5**
	Spacings	3.8*	0.2	1.5	0.4	0.1
Coefficient of variation (%)		34.9	23.4	11.3	13.9	32.6

*, **, denotes differences significant at P <5%, 1% respectively

4.3.4 Effect of seedling spacing on the total number of soil-inhabiting pests in the crop.

For each pest the number per seedling root was multiplied by the number of seedlings per acre at each of the respective seedling spacings to obtain a figure for the relative number of pests per unit area in the crop. The points were plotted from data obtained by taking a mean figure from four sites for each of the pests (Appendix Table 1). It can be seen from the points plotted in Fig.19 that close spacing results in an increase in the total numbers of all three pests per acre. The Collembola population estimate follows closely the ratios of seedling populations at the three spacings since there was, on average, similar numbers per seedling root. Millipede and particularly pygmy beetle numbers per acre are comparatively fewer when seedlings are closely spaced, than Collembola.

4.3.5 The effect of seedling spacing on root damage

The root damage scores, assessment of root damage and percentage of seedlings showing damage was analysed by Analysis of Variance. The effect of seedling spacing on these variates is shown in Table 20. At Bottisham and Shouldham the pest damage was due almost entirely to blaniulid millipedes; at Stalham to Collembola (Onychiurus spp.); at Melbourn to wireworms and at Magdalen and Broom's Barn to pygmy beetle. In most samples the seedlings from the 9 in. spacing had the most root damage. At Bottisham the later sample showed that the damage to the roots had increased and differences were often highly significant between spacings. At Stalham similar consecutive samples showed an increase in root damage but

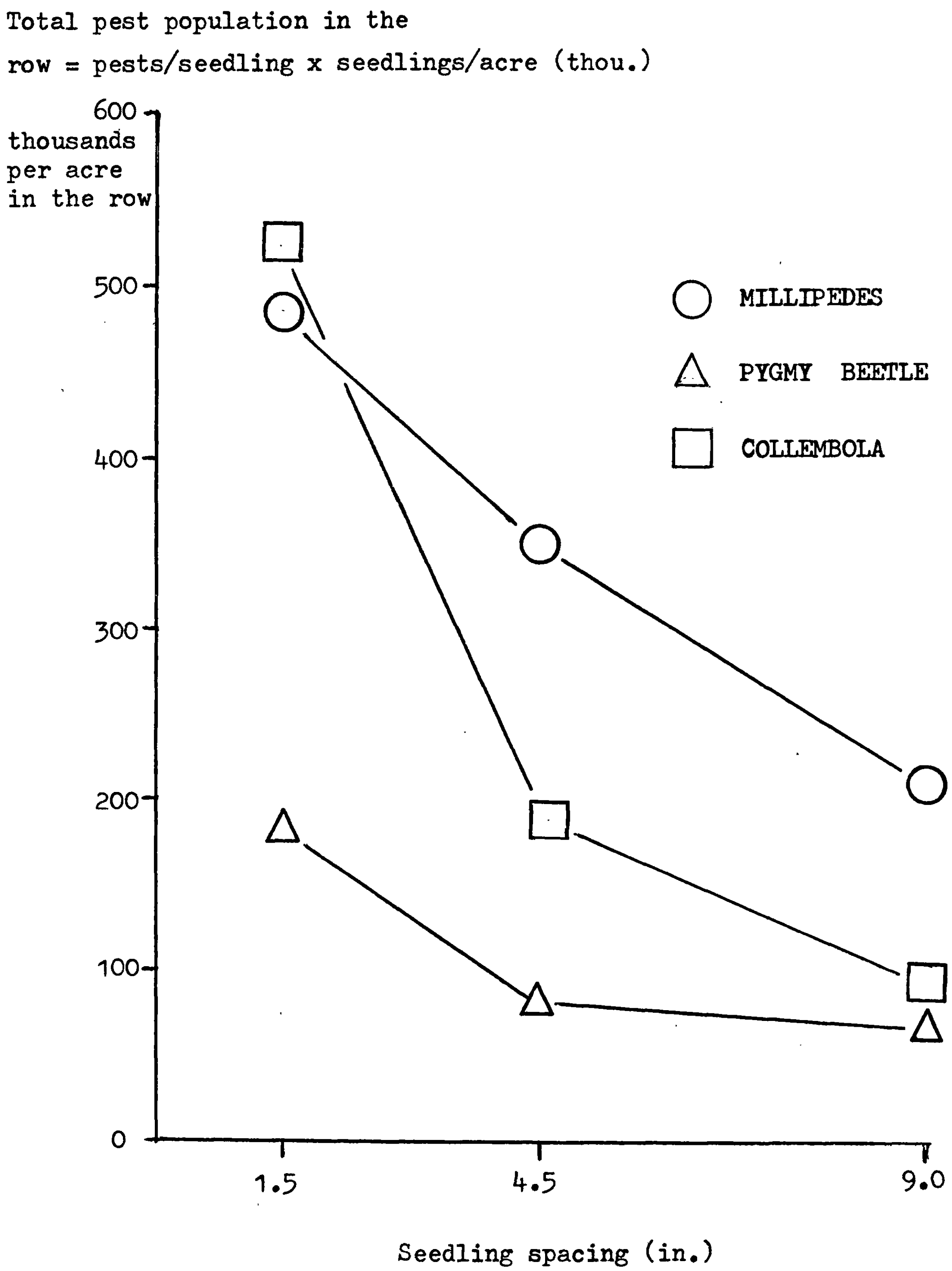


FIG. 19. <u>EFFECT OF SEEDLING SPACING ON THE TOTAL NUMBERS OF SOIL-INHABITING PESTS IN THE CROP.</u>

TABLE 20. EFFECT OF SEEDLING SPACING ON DAMAGE TO THEIR ROOTS BY SOIL-INHABITING PESTS

Seedling spacing (in.)	Mean pest damage score per seedling (0 - 5)							PMB* bites per seedling	% seedlings with root damage	
	Magdalen 5.6.72	Melbourn 18.5.72	Shouldham 1.6.72	Bottisham 14.5.74	Bottisham 20.5.74	Stalham 6.5.74	Stalham 21.5.74	Broom's Barn 1971	Magdalen 1972	Melbourn 1972
a 1.5	1.25	0.14	1.96	1.24	1.41	1.73	1.96	2.97	80.9	10.1
b 4.5	1.47	0.35	2.19	1.45	1.93	1.61	1.84	3.06	83.3	19.4
c 9.0	1.54	0.39	1.68	1.81	2.03	1.72	2.23	2.69	89.6	22.9
L.S.D.										
P = 5%	0.23	0.11	0.82	0.20	0.20	0.40	0.26	1.38	9.8	5.7
P = 1%	-	0.15	-	0.27	0.27	-	0.35	-	-	7.9
P = 0.1%	-	0.20	-	0.36	0.38	-	0.49	-	-	4.0
P a-b	n.s.	***	n.s.	*	***	n.s.	n.s.	n.s.	n.s.	**
b-c	n.s.	n.s.	n.s.	**	n.s.	n.s.	**	n.s.	n.s.	n.s.
a-c	*	***	n.s.	***	***	n.s.	*	n.s.	n.s.	**

*, **, ***, denotes differences significant at P <5%, 1%, 0.1% respectively

* PMB is an abbreviation for pygmy mangold beetle (pygmy beetle).

differences were only significant in the second sample. Analysis of the number of bites per seedling by pygmy beetle at Broom's Barn in 1971 showed no effect of seedling spacing whilst the percentage of seedlings with root damage at Magdalen was greater at the 9 in. than the other spacings, but was not significant. At Melbourn root damage by wireworms occurred on a significantly higher percentage of seedlings at 9 in. and at 4.5 in. than at the 1.5 in. spacing.

The dry weight of seedlings is governed by rate of growth. Growth can be affected if the ability of the roots to absorb nutrients is impaired by pest damage, particularly damage to lateral roots and root hairs. The results in Fig.20 indicate that at all sites (except Shouldham) seedling dry weight is greater when they are grown at 1.5 in. than at 9 in. spacing. This was particularly evident in the samples taken at Bottisham in 1971 and 1974 on different fields but where blaniulid millipedes were damaging the seedling roots; the difference in weight of the seedlings at 1.5 in. and 9 in. at both sites was highly significant ($P < 1\%$). At Broom's Barn in 1971 and at Melbourn in 1972 seedling weights from the different spacings were not significantly different. At Shouldham in 1972, seedlings at 4.5 in. were significantly heavier than those at 1.5 in. spacing and at Stalham 1974, those at 4.5 in. were significantly heavier than those at 9 in. spacing. At Bottisham and Stalham in 1974 the lightest seedlings had the most root damage.

Seedling vigour is a subjective assessment of relative growth and is linked directly with dry weight of the seedlings. The differences in vigour score between seedlings assessed at Bottisham and Magdalen were highly significant between spacings (Table 21)

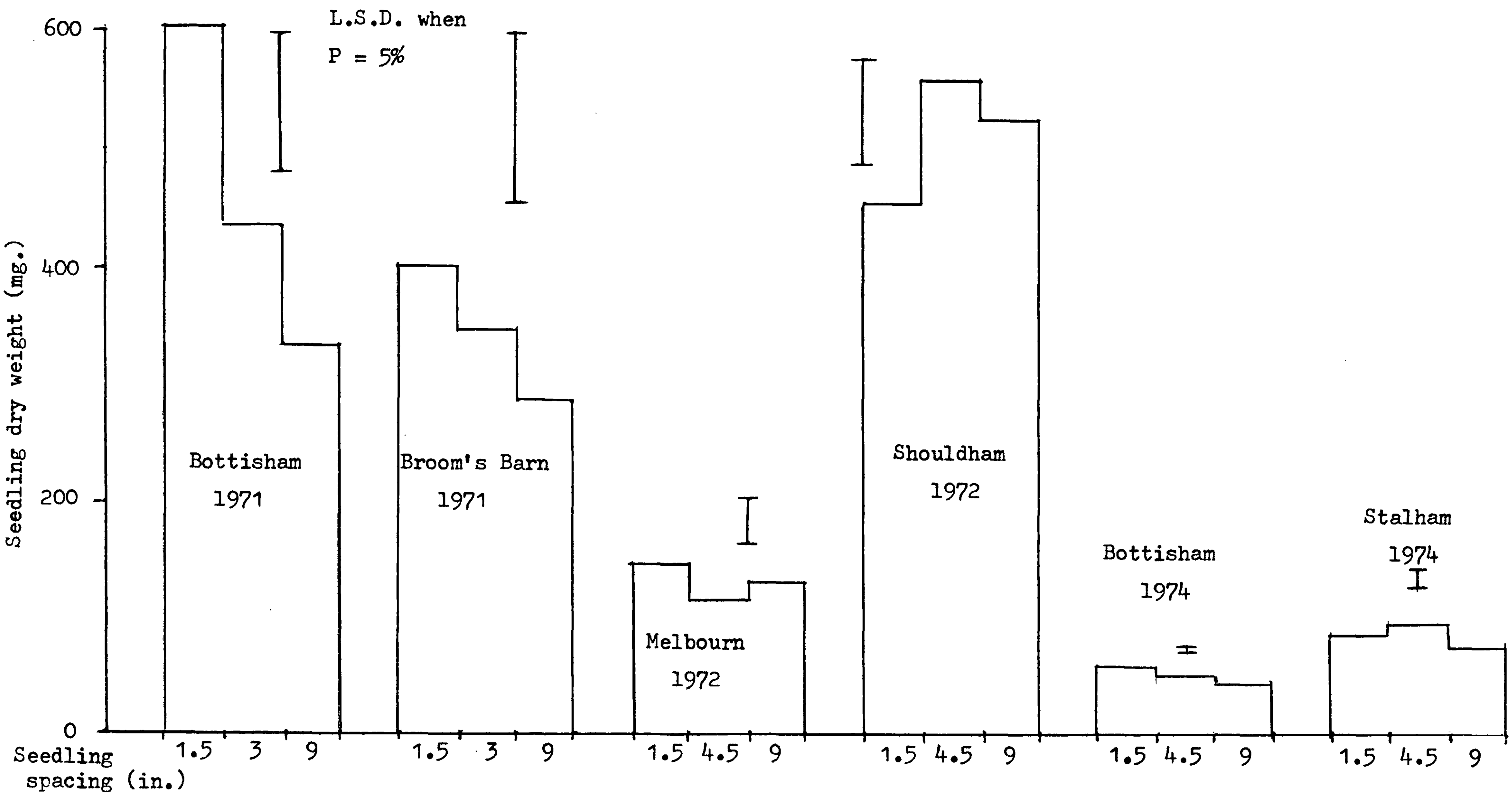


FIG. 20. EFFECT OF SEEDLING SPACING ON THEIR DRY WEIGHT

TABLE 21. EFFECT OF SEEDLING SPACING ON VIGOUR ASSESSMENT

Mean seedling vigour score (0 - 5)		
Seedling spacing (in.)	Bottisham 1971	Magdalen 1972
a 1.5	3.25	3.31
b 4.5 except 1 = 3.0	2.50	1.94
c 9.0	2.13	1.69
L.S.D.		
P = 5%*	0.37	0.51
P = 1%**	0.50	0.70
P = 0.1%***	0.70	0.97
P a-b	***	***
b-c	n.s.	n.s.
a-c	***	***

4.3.6 Effect of pre-emergence herbicide on pest distribution in the rows and the effect on root damage.

The data relating to the effect of herbicide on pest numbers in the root zone from seed spacing experiments was transformed to Log. (n + 1) and analysed by Analysis of Variance.

The number of pests in the root zone was not consistently higher where herbicide had been used but significantly more Collembola (Onychiuridae) were recorded around seedlings in treated rows at Broom's Barn in 1971 (Fig.21). At Shouldham more millipedes were found around seedling roots on herbicide-treated plots on both sampling dates (May and June) but at this site also numbers of pygmy beetle and Onychiuridae were fewer on treated rows; none of the differences were significant. There is no comparison of the numbers of each pest in and between rows, on treated and untreated plots owing to the high proportion of the population aggregating in the rows; relatively few pests were found in the 'between rows' samples.

Root damage was not consistently more on herbicide treated plots and only at Shouldham was it significantly greater where herbicide was used (Table 22) but this was not reflected in a significant increase in the percentage of seedlings with root damage at this or other sites (Table 23).

At all sites seedlings sampled from plots not receiving herbicide were consistently heavier than those from herbicide treated plots. Fig.22 shows the effect of herbicide on seedling dry weight at five sites; however, only at one site, Broom's

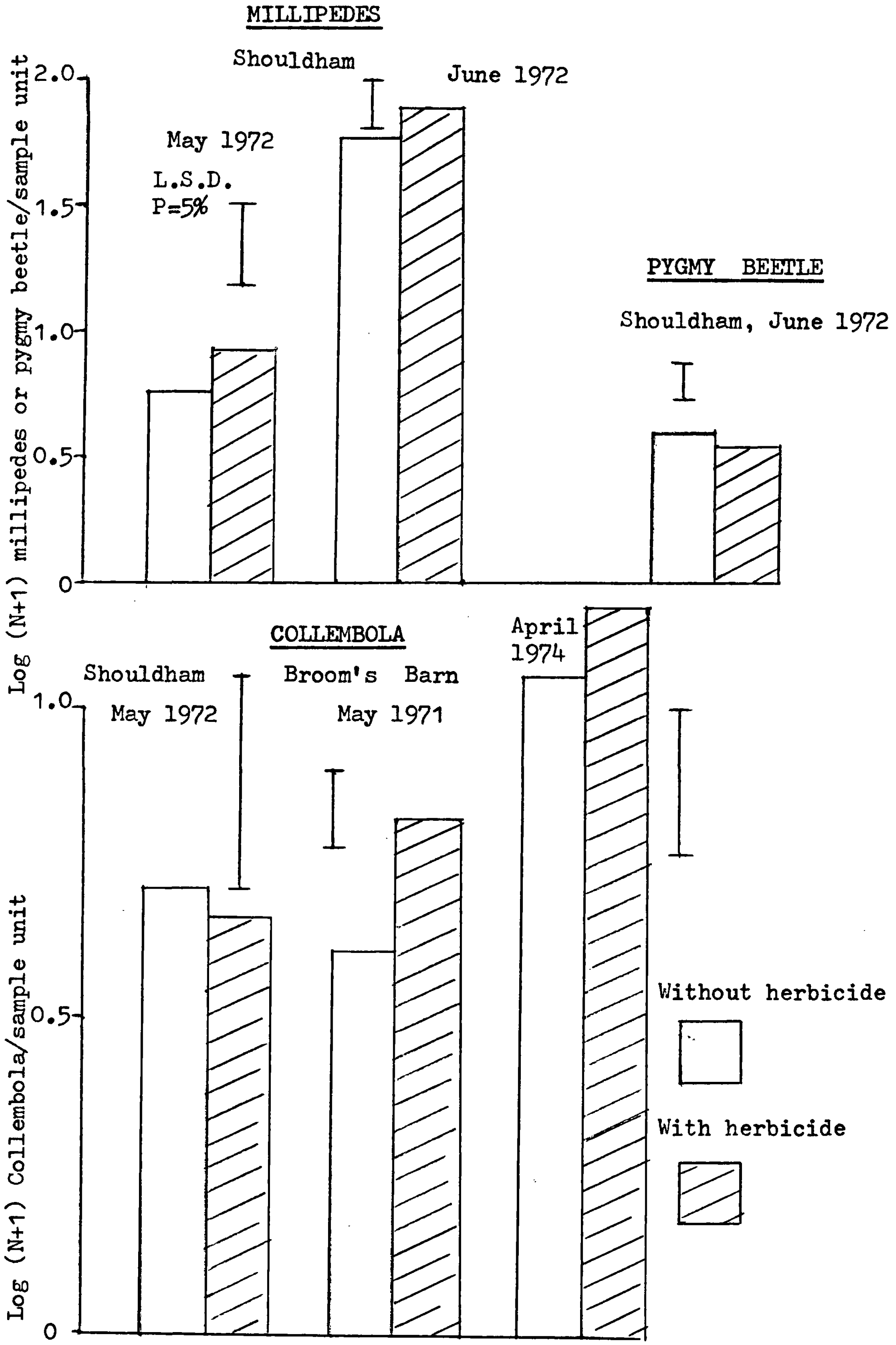


FIG. 21. NUMBERS OF PESTS IN THE ROOT ZONE OF SUGAR-BEET SEEDLINGS ON PLOTS BOTH WITH AND WITHOUT PRE-EMERGENCE HERBICIDE.

Barn in 1972, was the weight difference significant.

TABLE 22. EFFECT OF HERBICIDE ON PEST DAMAGE TO SEEDLING ROOTS.

Mean root damage score (0-5) per seedling

Site / Treatment	Broom's Barn 1971	Magdalen 1972	Melbourn 1972	Shouldham 1972
No herbicide	3.20	1.40	0.29	2.31
Plus herbicide	2.91	1.44	0.29	1.57
L.S.D. P = 5%*	1.38	0.19	0.09	0.67
P	n.s.	n.s.	n.s.	*

TABLE 23. EFFECT OF HERBICIDE ON PERCENTAGE OF SEEDLINGS WITH ROOT DAMAGE

Seedlings with root damage (%)

Site / Treatment	Magdalen 1972	Melbourn 1972	Shouldham 1972
No herbicide	84.2	18.0	85.8
Plus herbicide	85.0	16.9	81.7
L.S.D. P = 5%*	8.0	4.7	10.4
P	n.s.	n.s.	n.s.

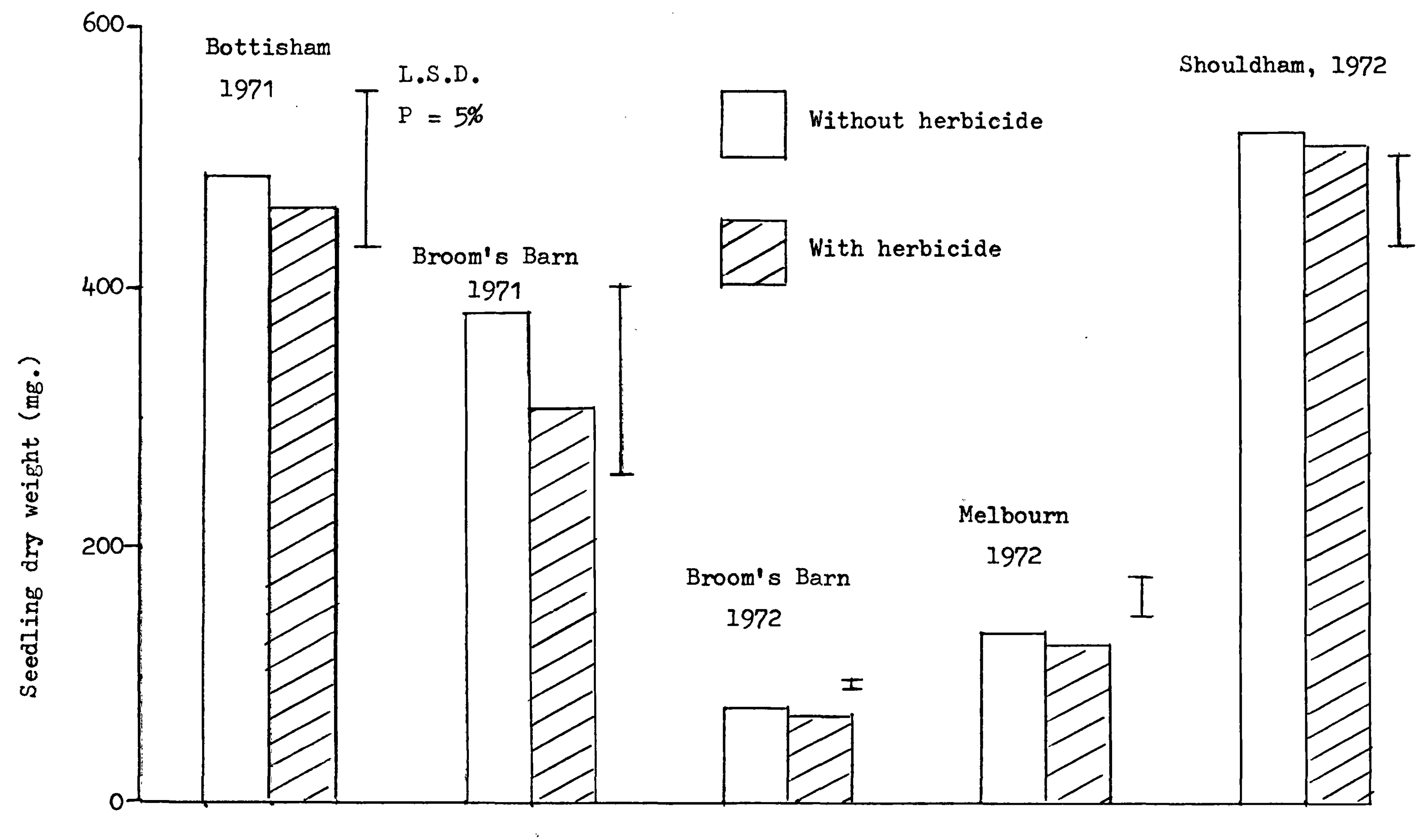


FIG. 22. EFFECT OF PRE-EMERGENCE HERBICIDE TREATMENT ON SEEDLING DRY WEIGHT.

4.4 DISCUSSION

The results show that blaniulid millipedes can be found deep in the soil in February and that surface samples to a depth of only 6 in. would give a misleading representation of the size of the population that was potentially available to infest the seedbed. Pierrard, Bonte and Baurant (1963) found Blaniulus guttulatus as deep as 80 cm. in the soil during December in France and Archeboreoiulus pallidus was found in greatest numbers in the 40-50 cm. stratum (Biernaux & Baurant, 1964a). Similarly, samples from Boston and Welney sites in 1971 showed that even Brachydesmus superus, normally considered as an active surface-inhabiting millipede species, can be found deep in the soil. It is possible that deep-ploughing carried the adults, usually present in large numbers in the autumn on the surface, down to the 4.7-8.7 in. stratum; Pygmy beetle was found at similar depth; ploughing would create more cracks to enable these essentially non-burrowing insects to move deeper into the soil profile in order to escape the winter climate.

Onychiurid Collembola were the most common soil-inhabiting arthropod in January and February and were often present in greatest numbers in the deeper samples; their numbers were also high in the seedbed when the crop was sown. In contrast, the presence of B.superus millipedes in autumn of the previous year or in early spring samples of the following year did not relate to their later population either quantitatively or qualitatively; as adults in early samples, only harmless eggs and young stages of B. superus were found later when the crop was sown.

The distribution of buried organic matter in the form of either ploughed-in cereal straw or a pig manure mixture may determine the depth distribution of pests in the soil. At a field at Seventh Drove (Cambs.) in February 1970, and at Welney in 1971, Brachydesmus was found immediately beneath the soil surface on cereal straw of the preceding crop. At Shouldham, in March 1972, blaniulid millipedes, mainly young stages, were found on a pig manure/straw mixture near the soil surface and at Kettering in early March 1973, the only Blaniulus were found in a core from 6-12 in. deep in the soil which included mainly compacted pieces of rotted cereal straw.

To a certain extent the distribution of Brachydesmus may follow that of old beet crowns since they are attracted to them as a food source, especially if they are in a rotted condition. Pygmy beetle can also be distributed according to food material. Examination of sugar-beet crowns from the preceding sugar-beet crop at Welney in March 1971 showed an average of 20 beetles per crown; highest numbers were recovered from those pieces which had the most prolific new shoot growth. These beetles form a reservoir of infestation that is readily available to attack the new crop of sugar-beet seedlings. Damage is always greatest and occurs earlier where the new crop is drilled on soil which was previously beet (Petherbridge & Stapley, 1935); however, a few pygmy beetle have been recovered from samples taken in winter and spring from fields where beet was not the preceding crop. On 29 November 1969, two separate fields at Stowbridge (Norfolk) were sampled to a depth of 6 in. with a 2 in. diam. auger. Of 5 samples taken from each field, where the preceding crop was potatoes and onions respectively, one beetle was recovered from each field. Beet had not been grown for up to 5 years in one field, but in each, the beet seedlings had suffered damage from pygmy beetle when sugar beet was last grown. At Kettering (Northants), samples taken to a depth of 12 in. in a field which had not been beet for 3 years returned one pygmy beetle per 16 samples taken in March 1973. It is possible therefore that some fields have a small resident population of pygmy beetle and one can but speculate that more will be present in fields which have a record of infestation by this pest when beet is grown.

The results do not show sufficient evidence that the population sizes of either pygmy beetle or millipedes in the seedbed can be accurately related to final seedling establishment but this may be related to the difficulty of obtaining an accurate estimate of the numbers of available pests. Those in the sample may be only a small proportion of those deeper in the soil which only extensive sampling will show. The proportion in the sampling zone may vary from site to site depending on soil characteristics such as texture and moisture, and perhaps distribution of localised food sources such as beet crowns. At those sites such as Terrington in 1973, and at Welney in 1971, numbers of pygmy beetle available were under-estimated and it is possible that samples were either not taken sufficiently deep or that most of the infestation was from flying beetles which arrived shortly after drilling.

For millipedes, which are true soil-inhabitants, the relationship was even more variable and additional explanations can be put forward.

The seedbed estimates of millipedes also included all juvenile stages and these are not capable of causing as much damage as the adult stages. Stadium II, the most numerous at Marham and Shouldham in the seedbed, could perhaps be omitted thus effectively lowering the size of the seedbed populations of pests at these sites. Particularly for millipedes, sampling to greater depths may give more meaningful relationships between population size and seedling establishment. At most sites where millipedes were numerous in the seedbed, an infestation by flying pygmy beetle followed in May or June and, depending on the size of the seedlings, could cause significant additional seedling loss. Soil sampling has shown that a pest complex exists in most soils with one pest usually dominant. The potential of Onychiurus as a pest in its own right has only recently been recognised and since this genus was present in varying numbers at most sites infested with either pygmy beetle or millipedes primarily then they may have contributed to the loss of seedlings to a greater or lesser extent.

Vertical displacement is a characteristic shown by most soil-inhabiting pests and the knowledge of the movements of pests in the soil, in particular the timing when most are in the top layer, can make assessment of populations both easier and more accurate. Finney (1941) obtained an approximate relationship between level of wireworm infestation and yield of oats by sampling mainly in the autumn or in late spring of the following year. It is probable that early spring (March or April) is too early for an accurate estimate based on sampling the top soil only, since migration towards the surface would not be complete and the results of sampling at sites such as Kettering would support this. Population estimates made in autumn of the preceding year could be useful since many pests have an autumn peak of numbers in the top soil. Biernaux (1968) found that blaniulid millipedes move back into the top soil in October with most in the top 10 cm. coincident with the remoistening of this layer by rain. Wireworms have also been found again in the top soil layers in autumn (Nadvornij, 1970). Soil samples taken later, in May and June, to estimate the numbers of the pests around seedling root zones show a clearer relationship with final seedling establishment. For pygmy beetle it forms an almost straight line relationship but for millipedes the data was insufficient. However, the results indicate that the presence of millipedes in the root zone does not necessarily mean a significant seedling loss, implying that those present are either incapable of inflicting serious damage or are not feeding.

Although sites were chosen for the highest probability of a pest attack, seedling establishment was sometimes of the order of 50-65% which is close to the national average of 60-65%. At Broom's Barn annual experiments from 1971-1973 recorded seedling establishments of 74, 94 and 76% for the three years respectively on plots without any insecticide treatment and in the absence of the usual soil-inhabiting pests. Onychiurus were probably present but any slight damage that may have occurred remained undetected. At none of the pest-infested sites did seedling establishment exceed 65% and it is possible that a proportion of the average 40-45% of the seeds that usually fail to produce a seedling may be due to Onychiurus damage when the seedlings are very small and not apparent to observers.

The distribution of soil-inhabiting animals in terms of both time and space is largely the outcome of vertical seasonal migrations or population displacements. The general tendency for many groups of soil animals is to live at greater depth in summer and winter than in spring and autumn (Dowdy, 1944). Distribution in time is important since it is useful to know when the animal is most common in the seedbed; this may be dependent both on time of breeding activity and on factors controlling vertical movements of adults and other stages in the soil. Distribution in space can be envisaged as the outcome of a redistribution mainly in the horizontal plane. Most true soil-inhabitants which feed on the roots of sugar-beet seedlings such as millipedes, wireworms, Collembola and Symphyla exhibit this upward movement in the soil whereas infestations of pygmy beetle are dependent more on the suitability of the conditions for flight from the fields of the previous year's crop where it overwinters on old beet crowns in the soil. The timing of this upward movement, bringing the pest into the seedbed in increasing numbers, will be dependent on both temperature and moisture preferences of each individual group and this will be reflected in the month when they are most numerous in the top soil where climatical changes exert most influence. Edwards (1958) reported on the vertical seasonal movements of Symphyla; Biernaux (1967a) and Glasgow (1939) have recorded the vertical movements of millipedes and Collembola respectively and associated them with temperature and moisture changes. Glasgow (1939) found greatest numbers of <u>Onychiurus ambulans</u> in winter and although he detected a general seasonal displacement, O. ambulans was always present in the top stratum (11.4 cm.) of soil throughout the year with peak numbers here in the March-April period. Heijbroek (1972), in Holland, found maximum numbers of Onychiurus in the top 3.5 cm. in April when the soil temperature was below 10°C but a few were present during March. Morris (1927) found that whereas O. armatus was common in the top 5 in. of bare arable soil in December, January and February at Rothamsted, the millipede B. guttulatus did not usually appear until March or April and were present in deeper samples. Biernaux & Baurant (1964a) in Belgium recorded blaniulid millipedes in the top 10 cm. in April of both 1962 and 1963 when the temperature at 10 cm. was between 5°C and 10°C but at this time most were deeper in the soil. Millipedes and Collembola are therefore active in the seedbed at similar low temperatures. Pierrard et al reported that B.guttulatus completely vacated the top 30 cm. of soil during December and January and did not appear in the top 10 cm. until the following March; numbers in this stratum peaked in May. From the literature it seems more likely that Onychiurus will be present in low numbers in the seedbed before blaniulid millipedes which have a more clearly defined migration and are absent from the top soil layers during winter. In the present study Onychiurus damaged seedlings at Broom's Barn and were found in large numbers in the root zone in March before millipedes were found around seedlings at other sites. Further evidence that Onychiurus is common in the seedbed in March was provided by soil sampling at the Kettering site in 1973 where there were an estimated 23.1 million Onychiurus but only 0.3 million blaniulid millipedes in the seedbed (0-10 cm.) at this time. Later samples in April showed six times as many

Onychiurus around the seedlings as millipedes but in May the numbers of millipedes had increased to equal that of the Collembola in the earlier sample. At Stalham, in 1973, soil samples, taken before drilling, on the 12th March and 20 March showed an estimated 7.4 and 8.0 million *Onychiurus* in the top 20 cm. of the seedbed and at the time of drilling there were 5.4 million in the top 10 cm. At most sites sampled from 1971 to 1974 *Onychiurus* was the most common arthropod in the seedbed before the crop was sown.

The numbers of pygmy beetle in the crop too, can vary greatly between successive samples even at sites where beet was grown for the second successive year. For sugar beet grown in the normal rotation numbers coming into the crop depend on the timing of its first flight from other fields but the factors controlling this are not understood. The size of the infestation may vary according to where the site is located, as the soil samples have shown, but the timing of the infestation determines the nature and severity of the damage; observations indicate that late infestations in June cause only minor foliage damage and the root remains largely unharmed. In Holland only late-sown sugar-beet crops suffer from damage from general migration by pygmy beetle but if good flight weather occurs exceptionally early then crops from normal sowings suffer (Heijbroek *in litt*.).

The results of the study show that, at least with millipedes, an increasing size of infestation of the top soil layers is accompanied by an increase in the proportion of the millipede population which moves towards the seedlings, indicating that the inter-row spaces are less attractive habitats than seedling roots. Michelbacher (1938) found a similar accumulation of the Symphyla, Scutigerella immaculata, in the top 6 in. in rows planted with snapdragons in a glasshouse in California; he found 13 times as many in the row as between the rows. Michelbacher (1939) also found greater numbers of S. immaculata in rows planted with sugar beet than in the inter-row spaces but the difference between the two became less as the sugar beet matured. Edwards (1961) reported that surface numbers of S. immaculata were very high within the rows where they were strongly attracted to young roots of tomato but not between the rows; a high between-row population was found only in deeper samples.

Millipedes, pygmy beetle and Collembola have been found to form distinctly aggregated distributions within the rows of seedlings or in bare soil; there were significantly more individuals in the row than between rows at the depth sampled. Healy (1967) found distinct aggregations of O. procampatus in spring and early summer coinciding with a period with larger numbers of juveniles but not with the period of maximum population size. The size of aggregations of O. armatus were considered by Glasgow (1939) to be 3-12 in. in diameter; aggregation in other Collembola was reported by Usher (1969). The ratio of sample variance to the sample mean has also been used by Blower (1970) to describe aggregation in Iulus and Polydesmus millipedes in a woodland habitat. Wireworms have also been found to have a non-random and aggregated distribution (Salt & Hollick, 1946). Changes in the density of the creatures in the soil often lead to apparent changes in their distribution pattern and we have seen that the degree of aggregation increases with density for millipedes, pygmy beetle and Collembola; the tendency for small populations to be less aggregated was also found with wireworms, (Finney 1941). Salt & Hollick (1946) observed changes in the distribution of wireworms with the age of the population. That non-randomness is probably the norm in true-soil-dwelling insects is suggested by the work of Salt, Hollick, Raw & Brian (1948) who found that it applied to nine groups of insects studied, including Symphyla, Collembola and Myriapoda.

The results show that of the three main pests studied, two, millipedes and pygmy beetle, can at some time distribute themselves around seedling roots in inverse proportion to seedling spacing (and density). Since there was no evidence that the numbers of Collembola in the root zone differed according to seedling spacing then there is probably an underlying fundamental behavioural difference from the other two pests. Millipedes and pygmy beetle will therefore be considered together when discussing the implications of this pattern. The evidence from sampling showed that any relationship between numbers per seedling and seedling spacing was one which depended on a suitable elapsed period between first and last sampling dates. Early samples showed comparatively low numbers in the seedbed and uniform distribution of both millipedes and pygmy beetle whereas later samples at the same site showed that some relationship between numbers and seedling spacing had developed, together with an increase in the number of pests in the root zone. The results suggest that the relationship is a function of the density of individuals in the seed bed. Sampling has shown that when present in relatively low numbers pests are less aggregated (more evenly dispersed); in addition,the sampling technique would be insufficiently sensitive to resolve small pest population differences between treatments. At relatively high densit~~y~~ies we have seen that aggregation is greater by virtue of an increase in sample variance and any differences that manifest between spacings may be insufficiently large to overcome the increase in sample variance. More accuracy would have been gained by processing cores taken around individual seedlings rather than by bulking together, thus increasing the number of degrees of freedom; however, the time necessary for the extraction and the seedlings available prohibited the adoption of this method of sampling. Although the bulking of cores tends to stabilize the 'between core' variation, the final mean figure per sample unit of 5, 8 or 10 sample cores is less meaningful than the mean number per core relating to one seedling, as in early samples. It was believed that the increase in the number of seedlings sampled compensated for the loss in accuracy in the statistical treatment of the results, which had the effect of making only relatively large differences significant.

The degree of 'clumping' in millipedes may be affected by their tendency to accumulate around previously damaged seedlings and leave some completely undamaged; this has been observed in laboratory feeding studies. Since damage retards seedling growth then undamaged seedlings will grow larger and in doing so will become more tolerant and resistant to attack. The damaged seedlings would consequently be attractive at some later time when millipedes had dispersed from the root zones of seedlings less attractive. It could be argued that this situation existed at the late-sampling times at the Kettering site when some overall dispersion away from the seedlings was apparent and differences in numbers of millipedes around differentially spaced seedlings was at a maximum. There was evidence that widely spaced seedlings are generally smaller (from dry weight analysis) than close-spaced seedlings. If increasing seedling maturity is associated with a reduction in susceptibility, and there is evidence for this, then this may provide a partial explanation for the pattern of millipede (and perhaps pygmy beetle) distribution on different spacings, viz: more per widely spaced seedlings. The amount of root damage was also greatest on widely spaced seedlings reflecting the increase in numbers of pests per seedling.

The decrease in root damage by pests on closely-spaced seedlings cannot entirely account for the increase in seedling size since at Broom's Barn sites, in 1971 and 1972, where damage by soil-inhabiting pests was very slight, the same clear relationship between seedling weight and spacing was evident. It is probable, therefore, that close-spacing is more conducive to rapid growth than wide spacing; if so, then it suggests the existence of some possible soil micro-climatical difference which may in turn affect the faunal composition in the root zone; the possibility that seedling spacing may affect the distribution of soil pests through differences in edaphic factors is not excluded. The effect of crop density on distribution of pests and damage has been studied in some vegetable crops. Wheatley & Hardman (1958) found that up to 17% more parsnips at the widest spacing were damaged than those on the closest spacing, but the latter were more heavily damaged. Similarly for carrots, Hardman & Wheatley, (1970) showed that those grown at the lowest density had the highest percentage of attacked plants. Numbers of the pest per unit area have also been found to be influenced; Hardman & Wheatley (1971) found that carrots grown at higher than normal density encourage carrot-fly populations to increase. In mini-cauliflowers, Thompson & Smith (1972) found twice as many cabbage root fly eggs per unit area on high density cauliflowers as around those conventionally spaced. In addition to differential attraction and shelter effects, some effect of host-plant density on the behaviour of natural enemies or incidence of disease was implicated in these findings.

The fact that closely spaced crops attract more soil pests per unit area illustrates a certain degree of specificity and the difficulty of locating those seedlings widely spaced, where millipedes, comparatively have more success.

With the almost universal use of pre-emergence herbicide in sugar beet to eliminate weed growth and the change in emphasis in pest control towards more long term use of cultural control measures, the suitability of weeds as alternative food needs investigation. Although the beneficial effects of herbicides are evident when weeds are large enough to compete directly with the sugar beet, little is known of the possible benefits of weed seedlings, particularly between the rows, in the early stages of seedling growth where a soil-inhabiting pest population is present. The present studies show no consistent effect of herbicide on numbers of pests in the root zone, but there was a general trend towards more pests in herbicide-treated rows. Weed seedlings collected from sites where millipedes have been numerous have often shown signs of pest damage to the roots. Plate 9 shows three weed seedlings which were growing alongside sugar-beet seedlings between the rows in soil infested with blaniulid millipedes, and earlier with *Onychiurus* spp. Each seedling has small lesions, particularly on the lateral roots and the centre seedling has a prominent hole in the cortex of the hypocotyl immediately below the soil surface. Since pygmy beetle were not present in the soil when the seedlings were collected, then the Collembola or millipedes are implicated. Although soil samples have not been taken centred over weed seedlings, as for sugar beet, no similar aggregates of soil-inhabiting pests have ever been observed around their roots, nor any evidence found of severe damage to their roots. Unlike sugar-beet seedlings, these weeds have a profuse growth of laterals and are better adapted to tolerate damage to their roots; damage to radicles of freshly-germinated weed seeds would be unnoticed in the field but some observations were made in the laboratory - *Boreoiulus tenuis* was found to cause damage to radicles of *Polygonum persicaria*, *P. aviculare* and *P. lapathirlium* immediately they emerged from the seed. The radicle tips of the latter were sometimes completely severed. The wide range of agricultural crops and fruit on which millipedes will feed is well documented (Rolfe, 1937-1939; Baurant, 1964) and the apparent susceptibility of sugar-beet seedlings to attack may be due partially to the efficient elimination of sources of alternative food by herbicides and to their predisposing effect to damage through their slightly adverse influence on the rate of early seedling growth.

PLATE 9. WEED SEEDLINGS FROM A FIELD WHERE BLANIULID MILLIPEDES WERE PREVALENT.

Pygmy beetles have been found to feed readily on a variety of weed seedlings in the laboratory including Chaenopodium album, Capsella-bursa-pastoris, Lolium italicum and Stellaria media, and also Rumex obtusifolius, Polygonum persicaria, P. convolvulus and Fumaria officinalis after a lapse of time (Petherbridge, 1941). This worker also took soil cores around beet seedlings, various weeds and bare soil and found that the beetles were mainly concentrated around the beet seedlings. Although significant differences between numbers found around the different weeds were not given, C. album was the most attractive weed in the two fields sampled.

Collembola, Onychiurus spp. have also been suspected of feeding on weed seedlings in the soil. Heijbroek (1972) found that where pre-emergence herbicides had not been applied, migration into the rows of beet was less pronounced and part of the Onychiurus population remained in the vicinity of the roots of weeds.

5. MILLIPEDES AS PESTS OF SUGAR-BEET SEEDLINGS

5.1. INTRODUCTION

Although some workers (Broleman,1920) have doubted that millipedes such as Blaniulus guttulatus can play a primary role in causing damage to crops, Brade-Birks (1930) and Rolfe (1937) have accumulated evidence that the families: Iulidae, Blaniulidae, and Polydesmidae are injurious to the roots of plants either in gardens or arable environments. Rolfe, emphasizing the importance of B.guttulatus, cites that of 130 cases of millipede damage reported by Advisory entomologists in England and Wales, 65 definitely referred to B.guttulatus (Cloudsley-Thompson,1950). Although some other species have been recognised as important pests, such as Orthomorpha gracilis, syn. Oxidus gracilus, (Edwards and Gunn,1961) in glasshouses, the blaniulids, in particular B.guttulatus and Archeboreoiulus pallidus, have been the most widely reported millipede pests in arable crops particularly in Belgium (Baurant,1964) where they are recognised pests of sugar-beet seedlings.

5.2. OBSERVATIONS

5.2.1. Symptoms of damage to seedlings by millipedes

Both field and laboratory observations were made of the most commonly occurring millipede species: Boreoiulus tenuis, B.guttulatus and Brachydesmus superus. Damaged seedlings were removed with a trowel from the field and brought back to the laboratory where the soil was washed from their roots and lesions inspected under a hand lens. The most typical case of damage by blaniulid millipedes is shown in plate 10. The root cortex of the seedling has been eaten in a peripheral pattern, approximately one inch below the soil surface and immediately above the region of the pellet leaving the inner vascular tissue as a thin central thread; it was very common to find seedlings almost completely severed at this point. Small lateral roots and root hairs are usually completely eaten. Such damage soon becomes blackened due to probable polyphenol-oxidase defence reactions and secondary invasion by fungi and bacteria. The foliage of seedlings damaged in such a way eventually shows a purple-yellowish discolouration which is due to the restriction of water and nutrient supplies to the tissues. The millipedes, which can be observed in the region of the damaged tussue, are usually found arranged in a radial pattern with their heads innermost. Plate 11 shows an aggregation of B.tenuis around a seedling root. The photograph was taken in the field (Shouldham, 1972) and the seedling is lying on its side with the foliage to the far right. A distinctive feature was the compact nature of the millipede aggregate.

In all field observations from 1970-1973 blaniulid millipedes were never found damaging seedlings before the middle of May and the most severe damage was evident in early June. In 1974, owing to drought conditions in early Spring, the millipedes were not seen until late June when the seedlings had already been singled. At this time damage by blaniulids was restricted to small lateral roots (Plate 12), particulatly those deeper in the soil; the damaged areas show as black and are restricted to the tips of lateral roots. Such damage would not seriously impair root growth although many millipedes may be found in the root zone.

PLATE 10. DAMAGE TO A SUGAR-BEET SEEDLING ROOT BY *BOREOIULUS TENUIS* x 4.

PLATE 11. AGGREGATION OF *BOREOIULUS TENUIS* AROUND THE ROOT OF A SUGAR-BEET SEEDLING

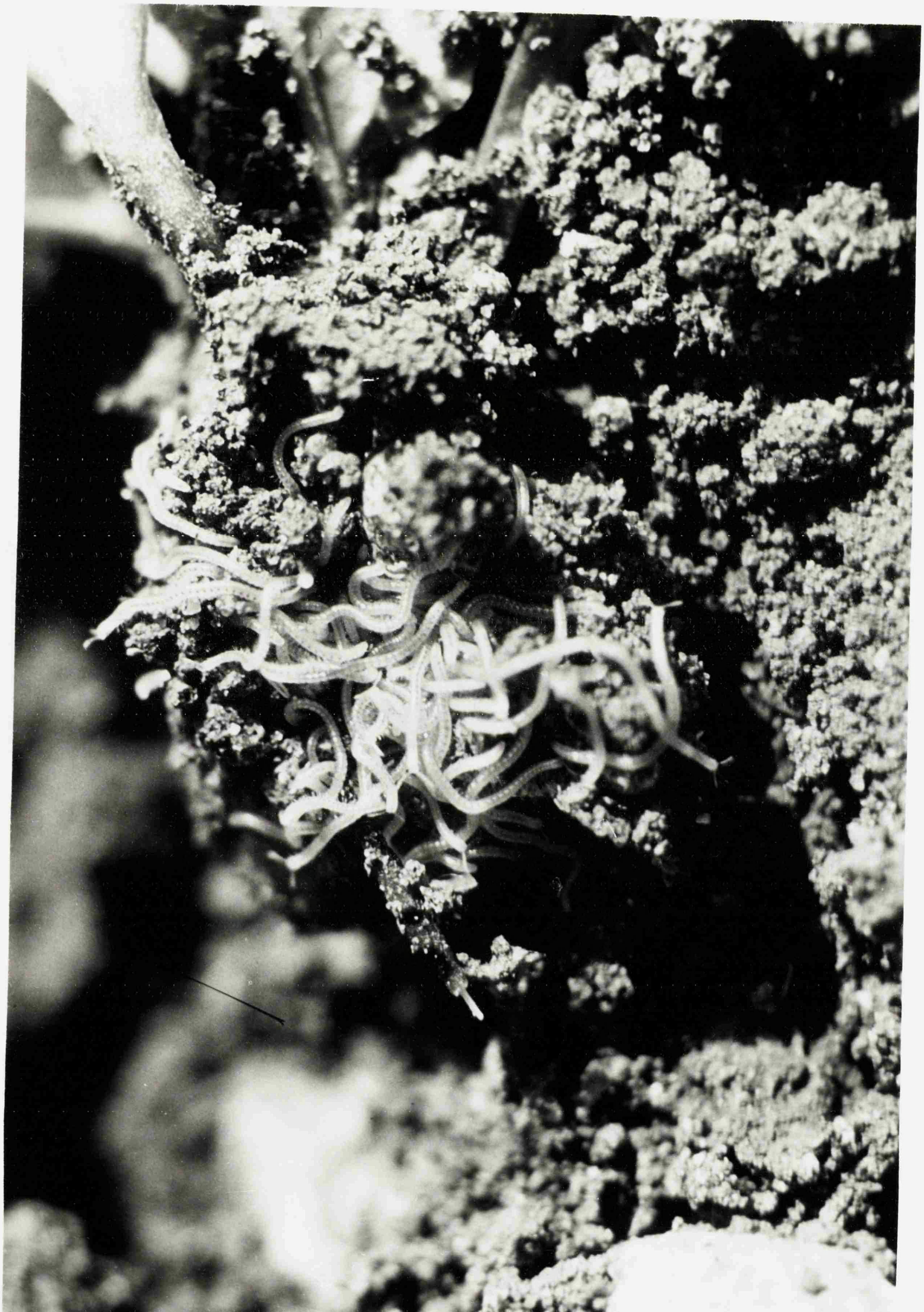


PLATE 12. DAMAGE TO THE LATERAL ROOTS OF AN ESTABLISHED SUGAR-BEET SEEDLING BY *BLANIULUS GUTTULATUS*

Polydesmids, Polydesmus spp. and B.superus, were rarely found in a stage suitable to cause damage to seedlings in the period April to May, although the adults were often numerous in the seedbed prior to drilling.

5.2.2. Damage to seedlings by millipedes in the laboratory

Millipedes were collected from the field and kept in sealed polythene boxes on moist soil at 5-7°C as a source of experimental material. Sugar-beet seeds, both raw and pelleted, were pre-germinated at laboratory temperatures until the radicle was visible. Approximately 5 seedswere placed in Petri dishes containing a layer of loosely packed moist soil together with 20 millipedes. The blaniulid millipedes quickly accumulated around the seed and damaged all parts of the young seedling, particularly the radicle and cotyledons, within 3 or 4 days in the laboratory. Plate 13 shows 3 B.tenuis millipedes around a germinated seed grown from a pellet; the cotyledons and radicle had been damaged and also the testa remaining in the fruit had been eaten. Plate 14 show a seedling which had been grown in soil in a box containing many B.tenuis collected from the site at Shouldham in May 1972. Although the cotyledons were hardly marked the radicle was extensively damaged and the lower half completely excised by continuous feeding lesions. Both seedlings shown would not have grown sufficiently for their cotyledons to have emerged above the soil surface under field conditions and this type of damage would usually remain unnoticed.

Brachydesmus did not cause such extensive damage to seedlings as the blaniulid millipedes, nor were as many ever attracted to any

PLATE 13. DAMAGE TO A GERMINATED SUGAR-BEET SEED IN THE LABORATORY BY *BOREOIULUS TENUIS*

PLATE 14. A SUGAR-BEET SEEDLING DAMAGED BY *BOREOIULUS TENUIS* IN THE LABORATORY

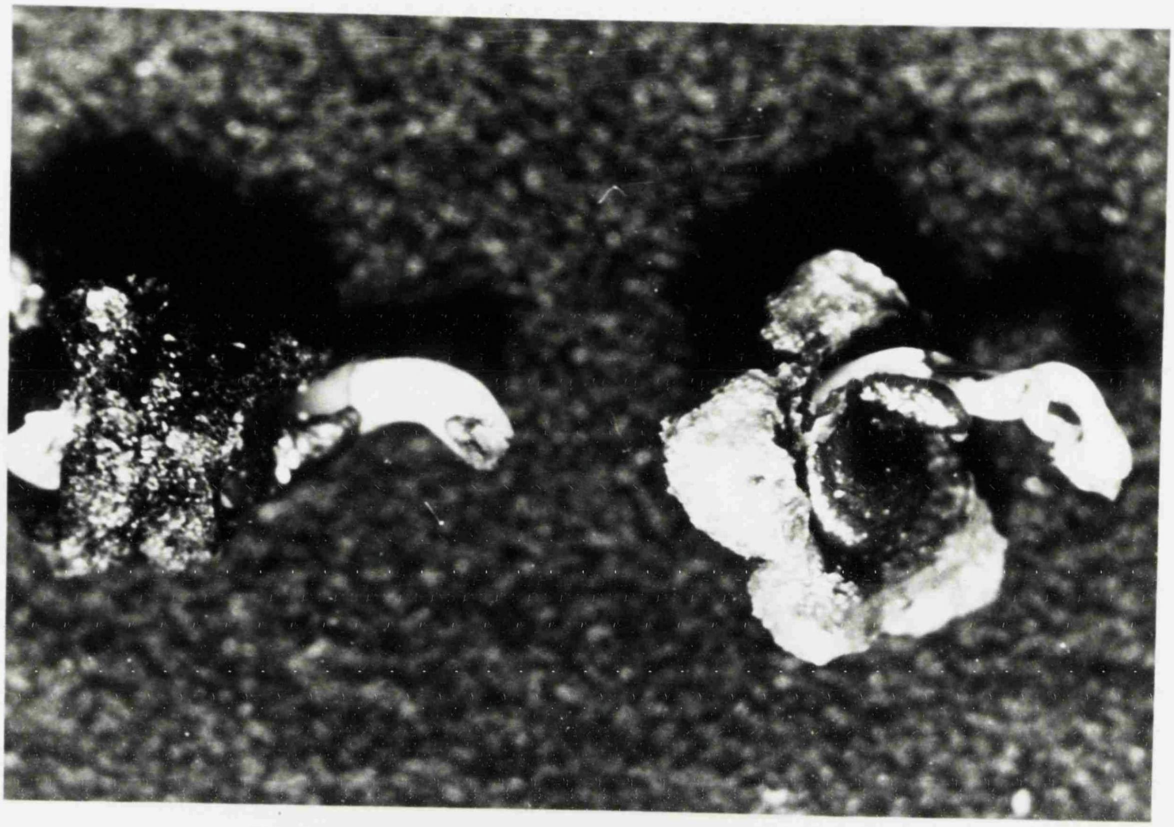
one seedling. Germinated seed were offered under the same conditions as for other millipedes but B.superus would damage only the radicle tips as they emerged from the fruit (Plate 15); such damage effectively inhibited further growth. Although stadium VII was sometimes capable of causing seedling death stadium V would cause only very small discrete lesions about the size of a pin prick and the seedlings would grow normally.

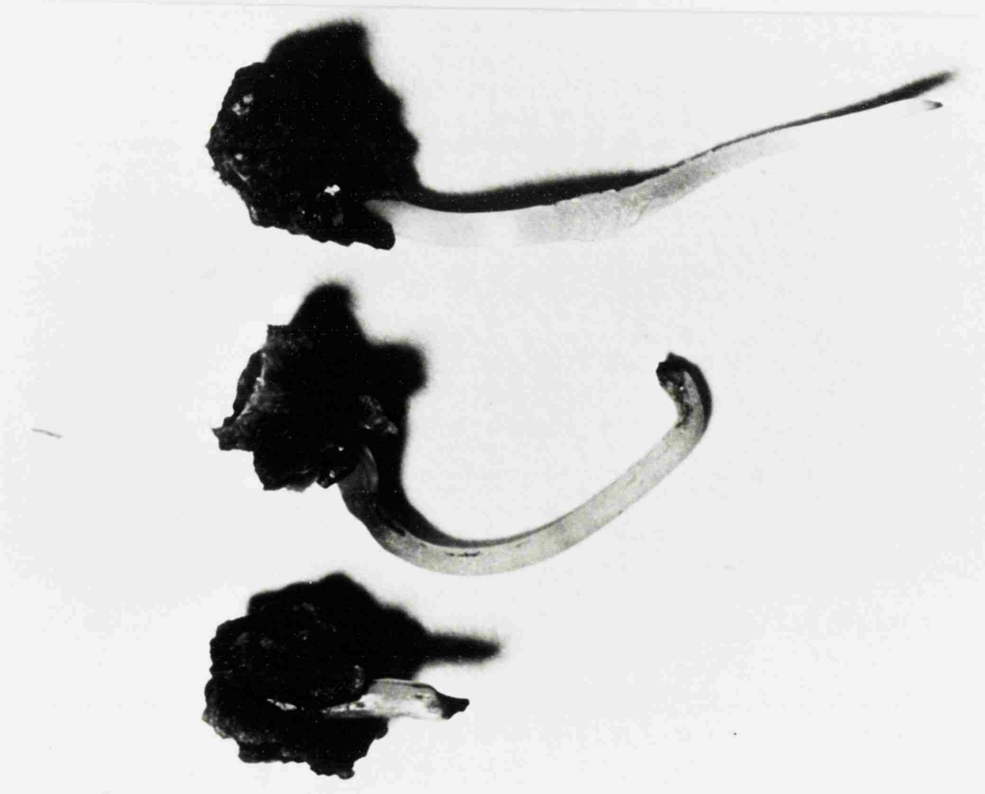
Six trials with B.guttulatus established conclusively that excised cotyledon material was more attractive than similarly excised fruit coat and testa, radicle tips, hypocotyl or seed pellet material. The cotyledons, which were cut from many seedlings, collected between 95 and 100% of the millipedes in the choice experiment in each trial and were usually almost completely ingested after 72 hrs.

PLATE 15. DAMAGE TO GERMINATED SUGAR-BEET SEED BY *BRACHYDESMUS SUPERUS* IN THE LABORATORY.

UPPER PHOTOGRAPH x 12

LOWER " x 5





5.3 DISCUSSION

It has been established that millipedes, pygmy beetle and Collembola are attracted to sugar-beet seedlings grown in the laboratory and are able to cause primary damage by feeding on the young roots. Whether this movement towards seedlings, which leads to aggregation around the roots, is the outcome of a random movement in the soil or a specific attraction in response to a root exudate, as for eelworms (Wallace, 1958), is not understood. Observations with a blaniulid millipede, in a Petri dish containing a single centrally placed germinated sugar-beet seed in the laboratory, suggest that there is no definite orientation but that the seed is located at random at distances of more than about 1 cm. At a radius of less than 1 cm there was some suggestion of an orientation towards the rhizosphere since the millipede passing within this area would rarely pass the seedling without stopping. In the field, random dispersal is probably most important in location of seedlings and the most active pests will therefore have the greatest chance of finding seedlings and forming the largest aggregates around the roots.

It was evident from the large numbers of millipedes recorded around roots of seedlings in the field that it would not be possible for all to be feeding at one time. It is possible that some other attractive force may be operating to keep the millipedes together as a cohesive entity. A laboratory experiment using a Y-tube, with one arm containing an extract of a macerated B.superus, showed that the arm containing the millipede extract attracted 71% of the released individuals, indicating that odour may play a part in the detection of millipedes already feeding on a seedling root. Peitsalmi (1974) demonstrated that Proteroiulus fuscus (Am Stein) will show thigmokinetic behaviour, which took the form of distinct aggregations of individuals; its compaction was dependent on air humidity. Although this worker could not explain how aggregations form it was suggested that the millipedes' olfactory sense could be involved. In the present study millipedes formed feeding aggregates around discs of water-agar in which dilute solutions of glucose or sucrose had been mixed; millipedes would also selectively feed on moist filter paper where individual raw sugar-beet seed had been left to germinate, indicating an attraction to leachable materials from the 'seed'. The nature of this material was not identified nor were any investigations carried out to identify the compounds exuded from the seedling roots into the rhizosphere. The range of sugars reported in exudates and the plants from which they were released indicates that exudation of sugars is a general phenomenon. Roots of oats, grown aseptically in nutrient solution, have been found to be covered with a mucilaginous oligosacharide which becomes densely colonized by micro-organisms (Rovira, 1965). The metabolic products from fungi and bacteria feeding on the root exudates may themselves serve as attractive media. Heijbroek (in litt.) found that Onychiurus is attracted to carbon dioxide sources in the soil; molasses at 400 l/ha sprayed between the rows decreased numbers in the row and increased numbers between the rows compared with the unsprayed control. Larvae of Otiorrhynchus, Melolontha and Agriotes are also attracted to concentrations of this gas produced by plant roots (Klinger, 1957, 1958). Mites and Collembola were considered by Moursi (cited by Kevan, 1965) to favour carbon dioxide and nitrogen concentrations.

The exudate theory may explain the localization of damge at or near the tips of seedling roots in the root hair and elongation zones where metabolism is most active but the frequent attack by millipedes on the delicate unfurling cotyledons as they emerge from the fruit suggests that texture and palatability may also be important in determining where damage occurs.

6. SOME ASPECTS OF THE BIOLOGY OF MILLIPEDES

6.1. STUDIES ON BRACHYDESMUS SUPERUS

6.1.1. INTRODUCTION

Brachydesmus superus Latzel ranks as the fifth most common millipede in the British Isles (Blower, 1972); it belongs to the order Polydesmoidae, Pocock, which are surface-dwelling and differs in being the only polydesmid habitually found in the soil. Most polydesmids have been suspected of causing crop damage (Rolfe, 1937). Brachydesmus differs in important aspects from the other polydesmids in having only 19 segments in the adult instead of 20; having 7 instead of 8 stadia and completing its life cycle in one year (Stephenson, 1961), compared with two or three years for most Polydesmus spp. (Blower, in litt.). This latter aspect is of considerable ecological importance since the more rapid increase in population size possible in Brachydesmus makes it of special economic interest.

The object of the study was to follow the development of B.superus stadia in the field and to note particularly the time when adults appeared in the soil. Since sugar beet is susceptible to damage by soil-inhabiting pests for only a limited period, of perhaps 2-4 weeks following the sowing of the seed, then the time of year that infestations develop are important since it will determine whether the seedlings become damaged by millipedes.

The effects of temperature on the rate of development of B.superus was studied in the laboratory and related to field observations.

6.1.2. Materials and Methods

Preliminary observations on the general aspects of the life-history of B.superus were made in the laboratory. The millipedes were collected from the field in autumn and observations made on pairing and egg laying at a temperature of 3.5-4^{o}C which approximated to the temperature of the millipedes' natural habitat. The millipedes were usually kept in glass Petri dishes having a layer of compacted moist soil and a lid.

The development of B.superus under field conditions was monitored by taking soil samples at intervals in spring and autumn in fields at Benwick (Cambs.), Boston (Lincs.) and Welney (Norfolk) where B.superus was known to be common. Samples were taken either with a spade or soil auger to a depth of about 6 in. and the millipedes were extracted either by hand-sorting or flotation using petroleum spirit. The latter method was the one adopted in the spring when a soil sampling programme was undertaken on experimental plots at some sites. The number of individuals at each stage of development from egg to the adult (stadium VII) were recorded.

At Welney, the millipede population in a single field was observed over a period of two years when sugar beet was grown for the second consecutive year.

Since temperature is the most likely factor controlling the rate of development in the field some laboratory observations were made of its effects on egg-laying and incubation in addition to observations of longer duration on the effect of temperature on the development of the larval stadia.

Covered crystalizing dishes containing moistened compacted peat soil with 10 adult female B.superus, collected from the field in February, were kept in the dark at five different temperatures. Two dishes (containing a total of 20 millipedes) were each kept at the following regimes: 2-3°C (incubator); 6-7°C (incubator); 5-11°C, (unheated room); 15°C, (incubator); 15-20°C, (laboratory). The millipedes were observed at intervals and dates of oviposition and the period of incubation for each egg batch were recorded.

B.superus, found in the adult stage only, were collected from a field at Welney in February 1971 where they were clustered around ploughed-in cereal stubble. 50 individuals (25 males and 25 females) were placed in 3 polythene boxes (7.1 x 4.7 x 1.8 in.) with tight fitting lids and kept at the following temperatures: in an incubator at 4°C; at laboratory temperatures (approx. 15°C-20°C); in an unheated room where the temperature varied from 5-15°C.

The rates of development of B.superus in each of the three different environments, as well as in the field, was monitored at intervals so that a comparison could be made. A random sample of soil (2 x 1.4 x 1.2 in.) was removed at intervals from each box and the number and percentage of millipedes in each stadium was recorded.

6.1.3 RESULTS

6.1.3.1. PRELIMINARY OBSERVATIONS

Adult millipedes collected from the field on December 16th 1970 constructed nests and oviposited in glass Petri dishes on January 8th and 12th when kept at 3-$4^{\circ}C$. Those collected from the field on January 13th, 21st and 27th and February 23rd and kept at $4^{\circ}C$, paired in March and April and oviposited in late March, April and May 1970. At this time all millipedes were kept at $4^{\circ}C$ since it was thought to approximate to the temperature in their natural surroundings. A check made in January 1970 in the field showed that soil temperature ranged from $6^{\circ}C$, at a depth of 3 in., to $3.5^{\circ}C$ at 12 in. when the air temperature was $9^{\circ}C$. At Nordelph (Norfolk) nests with egg clusters were found at ploughing depth on compacted soil in January 1970 when the above temperature data was collected.

Pairing between adult millipedes (plate 16) usually lasted between 24 and 48 hours and nest building commenced 7 to 32 days after pairing was first observed (average time for 11 couples being 14 days). Nest building procedure was similar for all adults when kept in soil. The female first made a small circular depression approximately 0.2 in. diameter on the soil surface (when it was compacted) or on the side of a lump of compacted soil beneath the soil surface; sometimes a nest would be contructed on the walls of the glass container. When the nest wall was approximately 0.1 in.

PLATE 16. UPPER - PAIRING BRACHYDESMUS SUPERUS ADULTS x 12.

LOWER - FEMALE B. SUPERUS IN THE FINAL STAGES OF NEST CONSTRUCTION x 16.

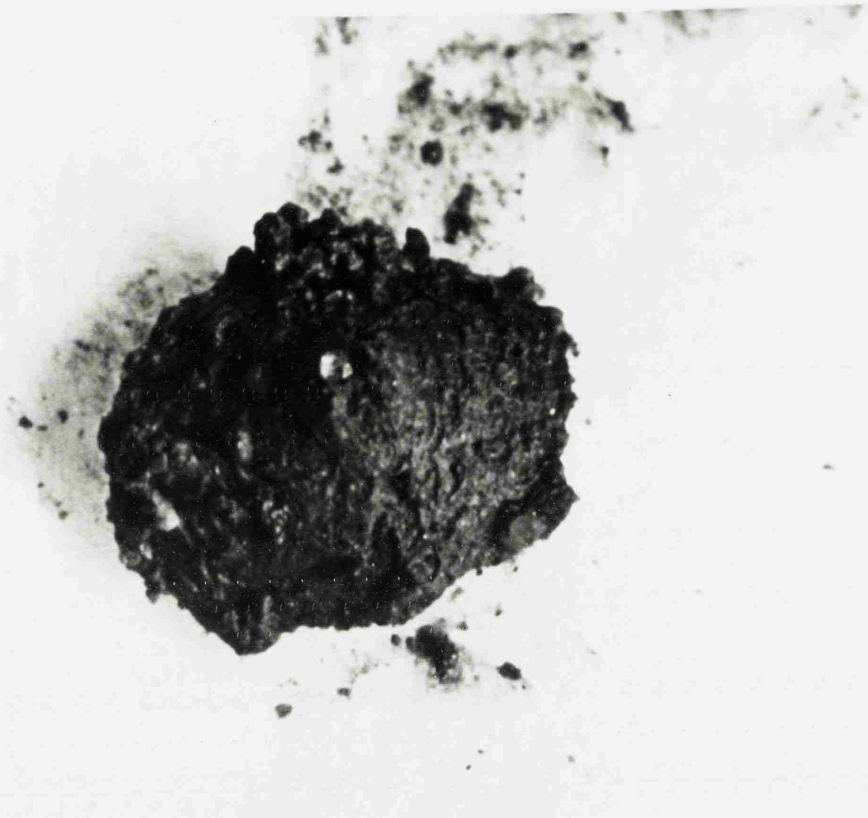
high, eggs were deposited as an egg mass within the nest. The nest walls and 'chimney' were then constructed thus completely enclosing the eggs (plates 16 and 17). The time taken for the complete nest building operation varied between 1 and 8 days for the 8 female millipedes under observation, the average time being 4 days, at 4°C; in the laboratory (approximately 15-20°C) it was often complete after 48 hours. Seven egg batches laid at 4°C were dissected and the number of eggs found varied from 29 to 89 per nest, the average number being 47 eggs. The female died shortly after reproduction; of 8 females observed the average life was only 38 days after pairing at 4°C. Many millipede pairs were observed to pair more than once. When adult millipedes were kept in potting compost only, eggs were deposited free within the medium and no nest was constructed. No eggs laid at 3-4°C hatched but those laid in the laboratory cultures (3 nests) took 15, 17 and 21 days for the emergence of the first stadia; this stadium always remained within the confines of the nest for at least 24 hours before breaking through the thin nest wall. Plate 18 shows stadium I and stadium VII B.superus.

6.1.3.2 SEASONAL VARIATION OF POPULATION AGE-STRUCTURE

Figs 23 and 24 show the change in age-structure of a population of B.superus under field conditions at Welney from February 1970 through to December 1971. In 1970 the adult stage was dominant until June and although a few eggs were found in the soil millipede feeding lesions were observed on the sugar-beet seedling roots. Samples in August and September showed a predominance of intermediate stadia; adults were few but they re-appeared in October in large numbers. Samples taken from the

PLATE 17. UPPER - NEST OF BRACHYDESMUS SUPERUS VIEWED FROM ABOVE x 15.

LOWER - EGG CLUSTER OF B. SUPERUS WITHIN THE NEST VIEWED FROM THE UNDERSIDE OF A GLASS PETRI DISH x 20.



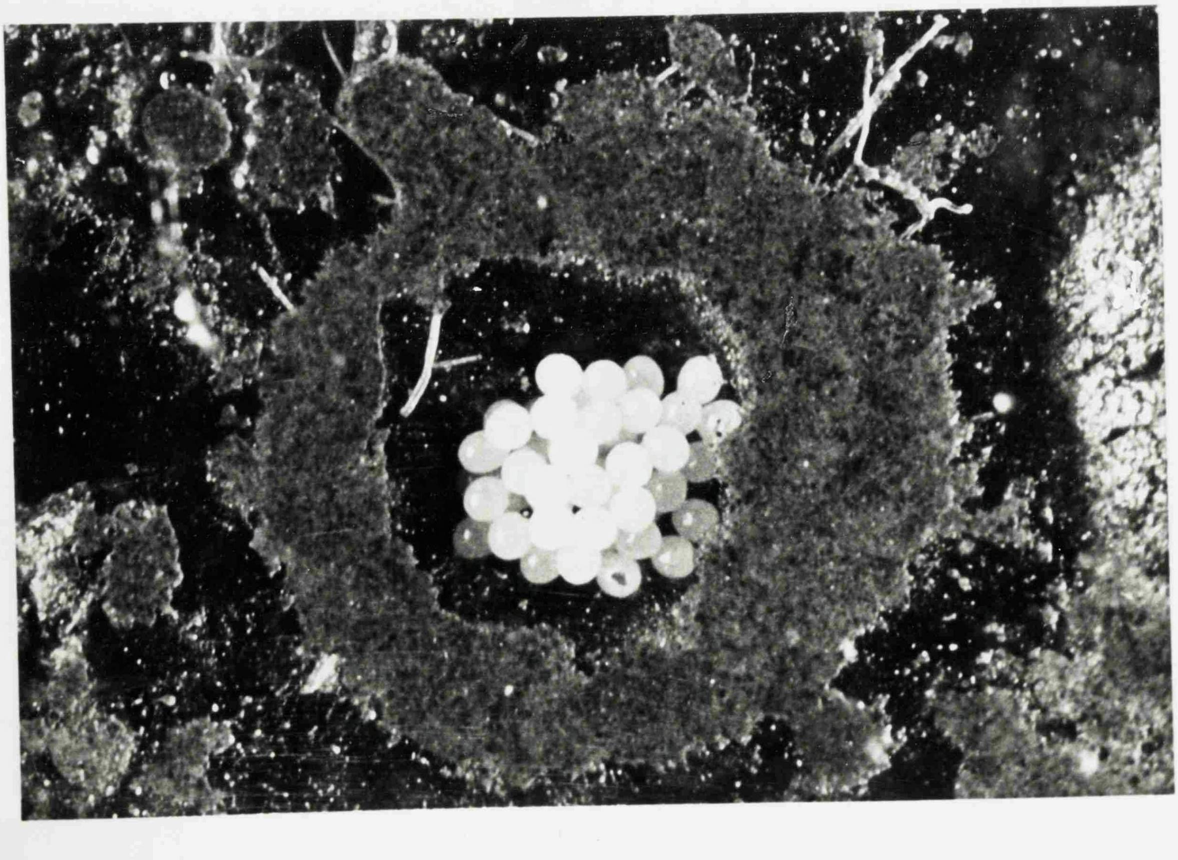


PLATE 18. UPPER - STADIUM I *BRACHYDESMUS SUPERUS* x 12.

LOWER - STADIUM VII *B. SUPERUS*, MALE x 11.

FIG. 23. POPULATION AGE-STRUCTURE OF BRACHYDESMUS SUPERUS AT WELNEY 1970.

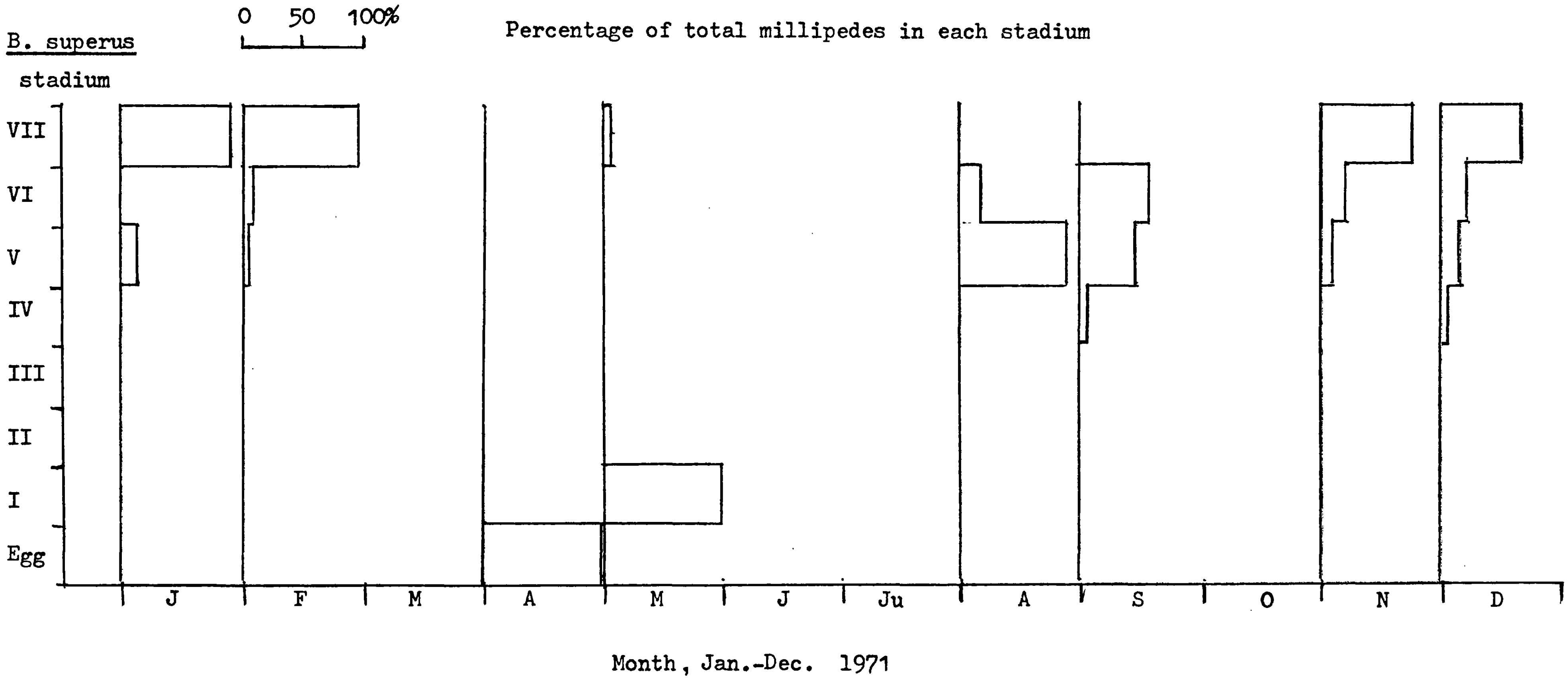


FIG. 24. POPULATION AGE-STRUCTURE OF BRACHYDESMUS SUPERUS AT WELNEY 1971

same area in 1971 contained mainly adults until February. No samples were taken in March, but in April and May, although the adult stage was rarely found, many eggs and stadium I were present in the top-soil; no millipede damage to seedlings was observed. By August and September stadia lV, V and Vl were common. Adults were present again in samples taken in November and December. At Boston soil samples taken in April and May 1971 on experimental plots showed a population of B.superus having a similar pattern of development with Stadium I predominating in May. In similar soil cores taken at Benwick in 1972 Stadium I was again more numerous than the adult stage in May (Fig 25).

6.1.3.3 EFFECT OF TEMPERATURE ON STADIA DEVELOPMENT

The millipedes made nests and laid eggs during March 1971 in the laboratory showing that pairing and fertilization had occurred previously in the field. Table 24 shows the treatments, dates of oviposition, number of nests constructed and the incubation period for the eggs at each temperature régime. No eggs laid at 2-3°C and 6-7°C hatched and the eggs were attacked and killed by fungal hyphae after eight weeks. At 5-11°C, 4 of the 5 egg masses hatched, taking 58-60 days. At 15°C and 15-20°C the average incubation periods were 26 and 21 days respectively.

Fig 26 shows the percentage of B.superus in each stadium on each sampling date for the three temperature régimes. At 4°C eggs and nests were produced in April and May but all adults had died by June and the eggs did not hatch. At 15°C-20°C, Stadia I, II and

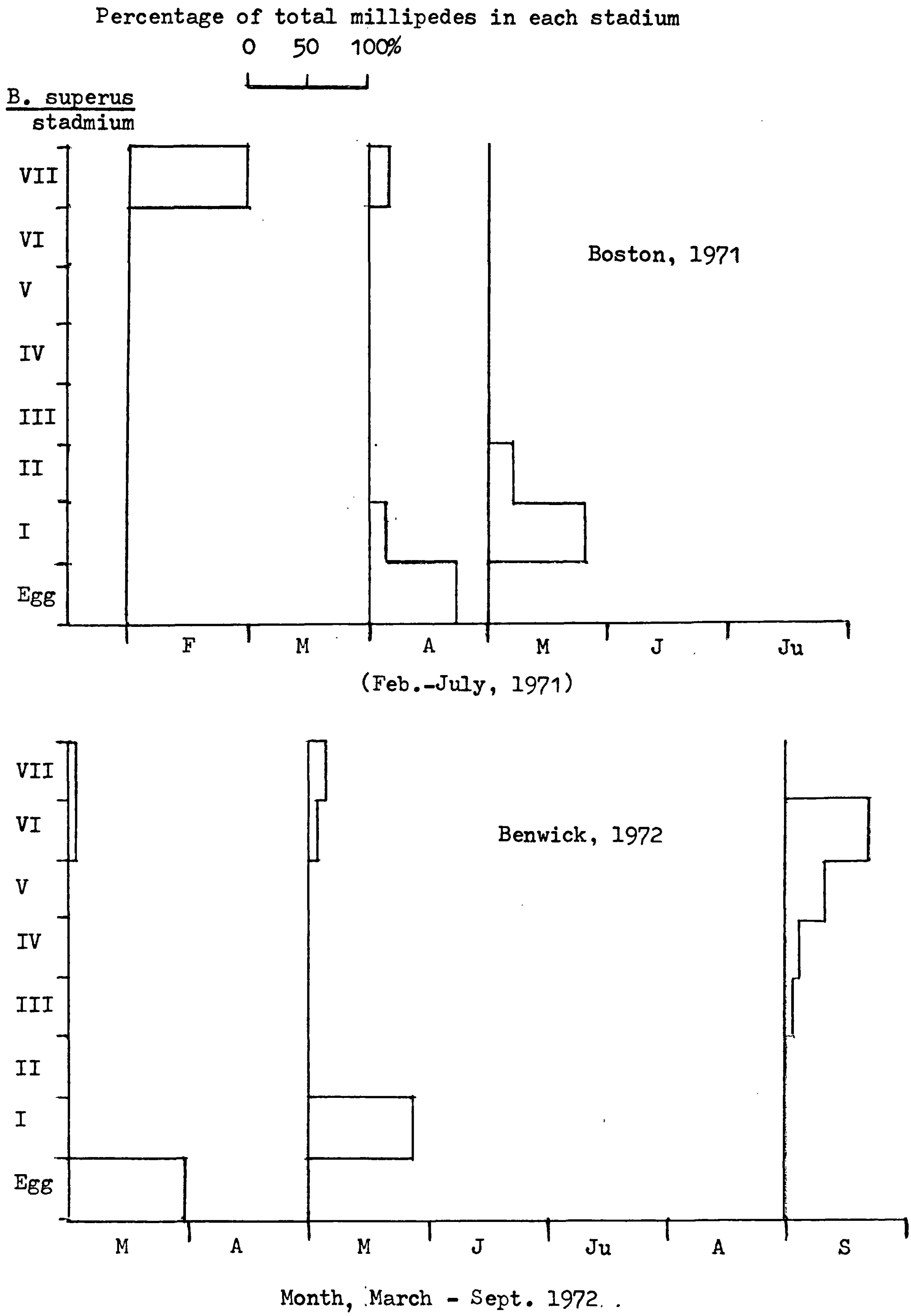


FIG. 25. POPULATION AGE-STRUCTURE OF BRACHYDESMUS SUPERUS AT TWO SITES IN 1971 AND 1972.

TABLE 24 EFFECT OF FIVE DIFFERENT TEMPERATURE RÉGIMES ON OVIPOSITION AND EGG DEVELOPMENT IN THE LABORATORY

Treatment	Temperature	Date of oviposition	Number of nests	Incubation period (days) for each egg mass
1	(2-3°C)	8.3.72	1	none hatched
		13.3.72	1	" "
2		9.3.72	1	" "
		10.3.72	1	" "
3	(6-7°C)	5.3.72	2	" "
		6.3.72	1	" "
		7.3.72	3	" "
		13.3.72	1	" "
4		5.3.72	1	" "
		6.3.72	1	" "
		12.3.72	1	" "
		13.3.72	1	" "
5	(5-11°C)	5.3.72	3	59, 59, 60
		6.3.72	1	none hatched
6		6.3.72	1	58
7	(15°C)	8.3.72	1	25
		12.3.72	1	26
		12.3.72	1	26
8		7.3.72	2	26, 26
		12.3.72	1	27
9	(15-20°C)	5.3.72	2	22, 23
		6.3.72	1	20
		7.3.72	1	20
		13.3.72	1	21
		18.3.72	1	none hatched
10		5.3.72	1	18
		10.3.72	1	21

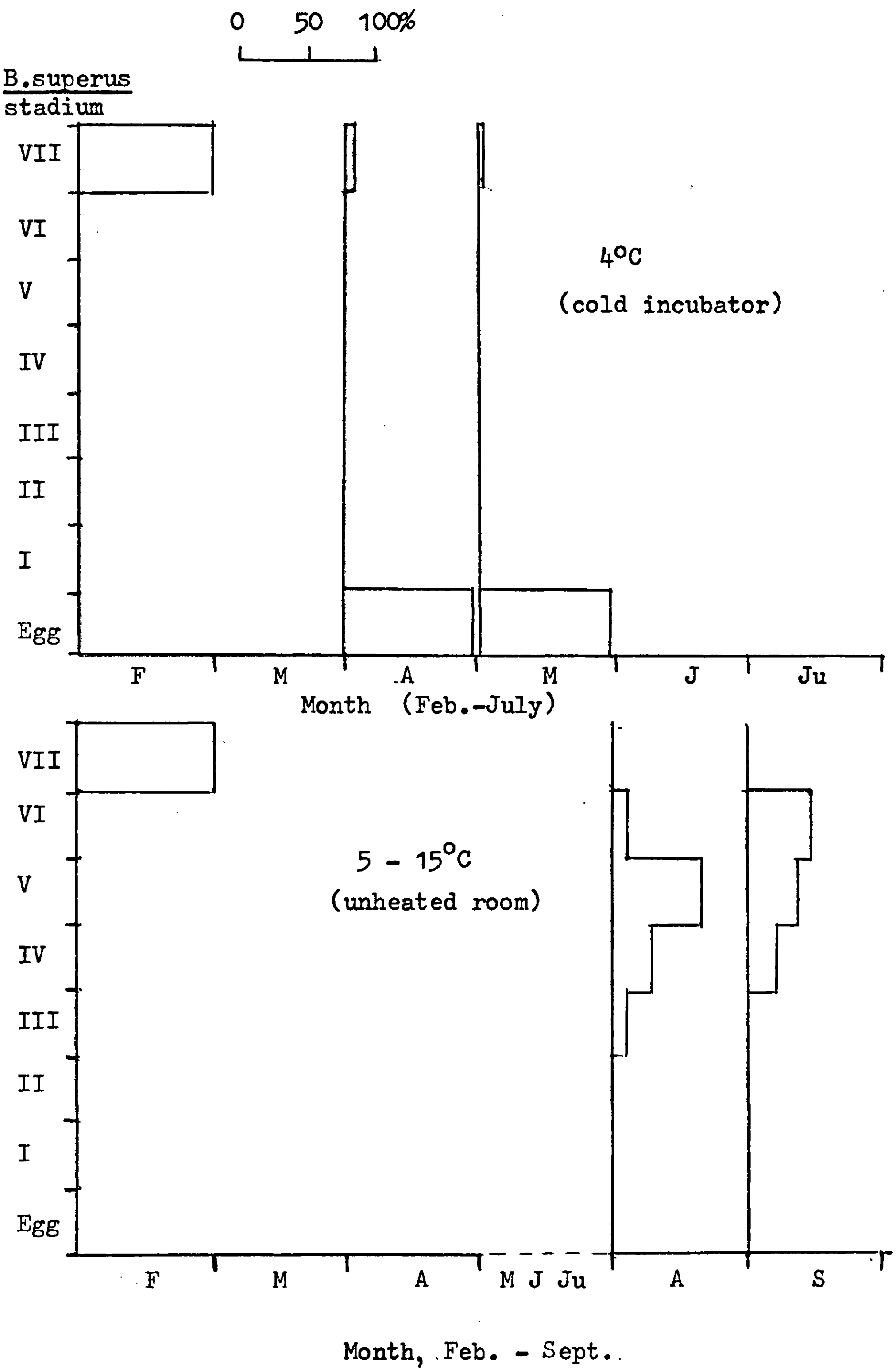


FIG. 26. STADIA DEVELOPMENT OF BRACHYDESMUS SUPERUS AT DIFFERENT TEMPERATURES. continued on P. 145

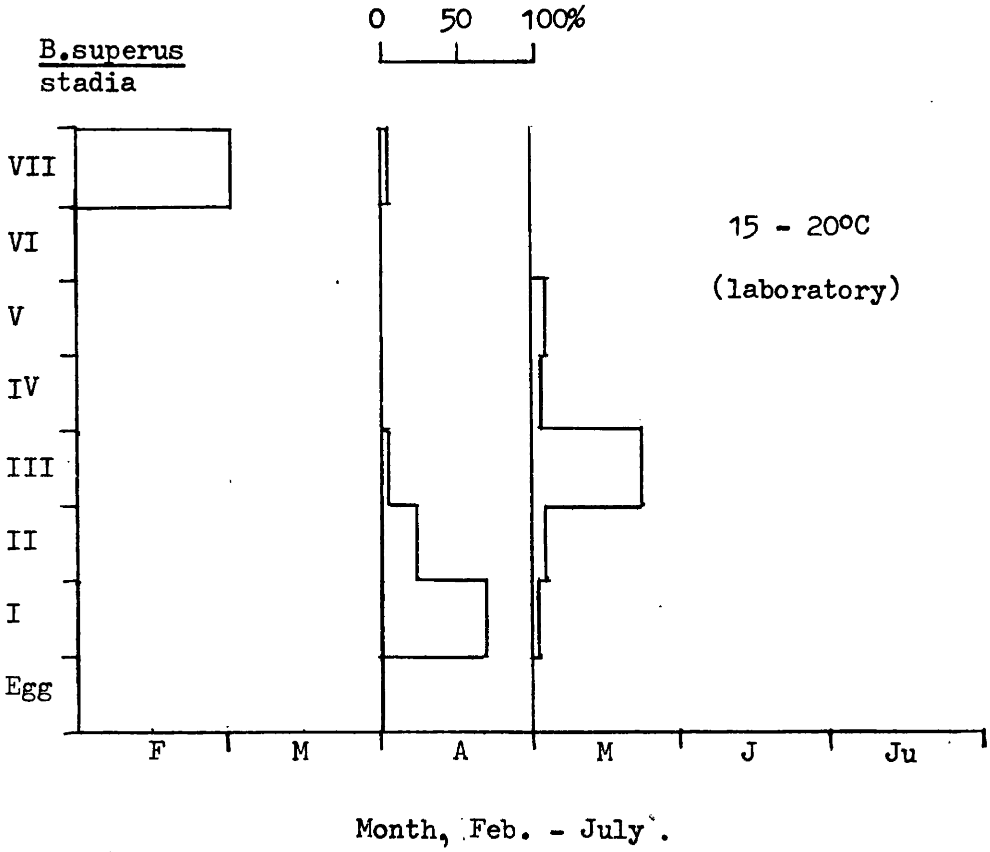


FIG. 26. (CONTINUED)

III were found in April and by May some had reached stadium V; all millipedes were dead by August. In the unheated environment, which more closely followed that in the soil, 63% were stadium V and some B.superus had attained stadium Vl by the end of August; this paralleled the rate of development at Welney where the adults were collected, here stadia V and Vl were abundant in late August and September 1971.

6.1.4. DISCUSSION

The results show that, although the rate of development of B.superus in the field can vary between two successive years, adults ~~are~~ were the overwintering stage with males predominating in both years and eggs hatch into stadium I during spring.

Stephenson (1961) gives a detailed account of the life cycle of B.superus and found that, of adults mated in Nov.-Dec., males died from December to January whilst the females overwintered and oviposited from March to June. Sexually immature 'intercalary' males had an additional moult, became sexually mature in January-April and mated with overwintering females from April to July; the females ovipositing from June to July. Neither of these interpretations fits the present data exactly. The predominance of males, especially in February 1971, would suggest a closer relationship with the second type of development but at Welney eggs and stadium I were found earlier, in April and May 1971. At the Boston site the occurrence of stadium I in April and May would suggest oviposition even earlier in that year, as at Benwick in 1972 where eggs were found in March. In the laboratory studies in 1971 there was evidence that the adults had mated prior to their collection from the field. The results show that females oviposit even at 4°C and thus some eggs may be deposited in the soil during winter as field observations confirm. Subsequent egg development and rate of development of stadia can be markedly affected by temperature. It is possible therefore that ambient temperature during winter and early spring can affect the

the rate of maturity of B.superus, development of eggs and appearance of young stages.

The unpredictability of B.superus as a pest is emphasised in the results of field sampling and that this may be due to variations in its habitat temperature from year to year is given some support.

6.2 STUDIES ON SOME BLANIULID MILLIPEDES

6.2.1. INTRODUCTION

The blaniulid millipedes *Archeboreoiulus pallidus*, *Blaniulus guttulatus* and *Boreoiulus tenuis* have been found in sugar-beet fields in spring and have been implicated in causing damage to young seedlings. The life-history of *B.guttulatus* has been described by Kinkel (1955) who collected specimens from agricultural land and cultured them in Petri dishes containing filter-paper and moistened Plaster of Paris. This worker also observed this species in the field, noting the breeding periods and appearance of the different stadia in Germany. Breny and Biernaux (1966) have also kept and observed *B.guttulatus* in culture and related their observations to data obtained as a result of soil sampling infested sugar-beet fields in Belgium throughout the year.

This section describes the culture of *B.guttulatus*, which has been attempted on the same lines as Kinkel (1955) and also describes some studies in the field on the life cycle of blaniulids, investigations of the seasonal variation in populations and their vertical distribution in the soil.

6.2.2. MATERIALS AND METHODS

6.2.2.1. RECOGNITION OF BLANIULID STADIA

Post embryonic development of blaniulids is anamorphic; there is a succession of larval stadia and at each moult there is an increase in number of body rings and pairs of legs. Since each of these body rings carries a pair of defence glands, each of which show as an orange or red spot, one on each side, then their number can be used to determine the stadium of the blaniulid. Stadia I - VI, the last juvenile stadium, can be determined accurately since no two stadia may have the same number of gland pairs. Maturity is achieved in stadium VII after which they may continue to moult, adding to their total number of glands and legs but numbers added at each moult may vary and hence result in an overlap in the number of these characters in each adult stadium. It was thus relatively easy to separate stadia I - VI by counting the number of pairs of defence glands. Stadium VII and older stadia were counted as 'adults'. The key provided by Brookes (1963) was used to separate stadia II - VII of both B.guttulatus and B.tenuis whilst Blower (1958) provided the key for each species. Both Kinkel (1955) and Biernaux (1968) described stadium I. Information on embryonic and post-embryonic development of B.guttulatus was given by Kinkel (1955). Characters used to separate the stadia are shown in table 25. The stadia were generally grouped into three categories: II, III - VI, VII +. Since it was very uncommon to find I then stadium II indicates the presence of the youngest stadia, III - VI are intermediate stadia and the last group includes all adults from stadium VII onwards. Some stadia are shown in plates 19 and 20.

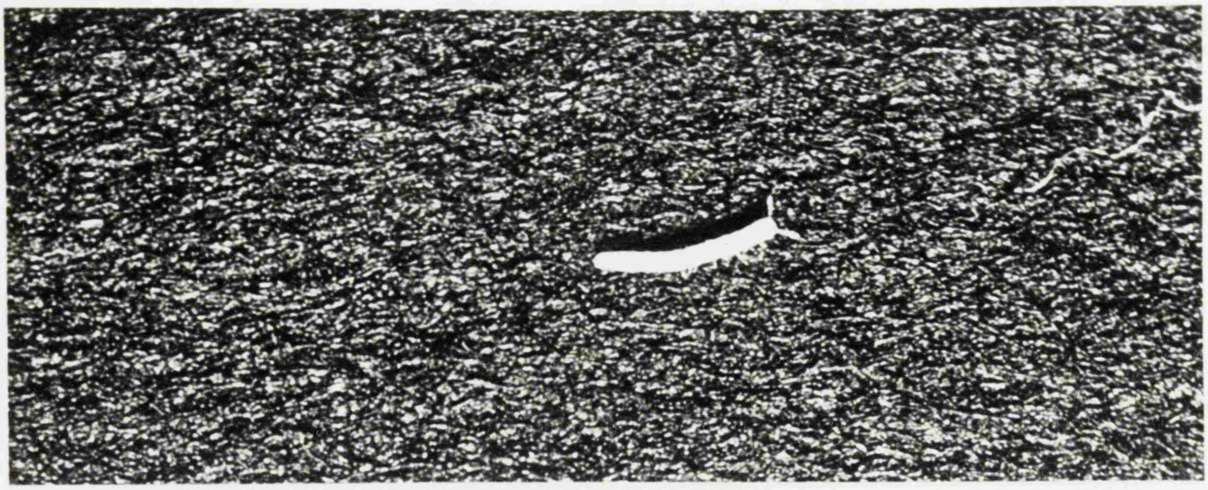
TABLE 25 CHARACTERS OF THE STADIA OF TWO BLANIULIDS FOUND IN SOIL SAMPLES

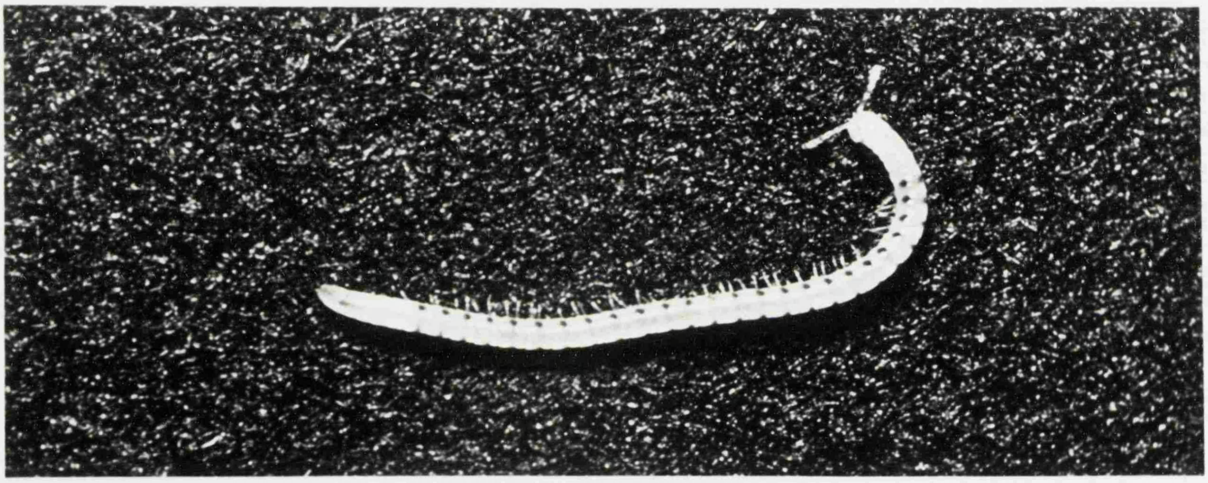
	Boreoiulus tenuis	Blaniulus guttulatus	Source of Information
Stadium I			
pairs of legs	-	3	(Kinkel, 1955)
No. of gland pairs	-	0	(Biernaux, 1968)
Stadium II			
pairs of legs	9	7	(Brookes, 1963)
No. of gland pairs	2	1	
Stadium III			
pairs of legs	17	17	"
No. of gland pairs	6	6	
Stadium IV			
pairs of legs	25-27	25-27	"
No. of gland pairs	10-11	10-11	
Stadium V			
pairs of legs	35-37	35-37	"
No. of gland pairs	15-16	15-16	
Stadium VI			
pairs of legs	43-47	43-49	"
No. of gland pairs	19-21	19-22	
Stadium VII			
pairs of legs	53-55	53-59	"
No. of gland pairs	24-25	24-27	

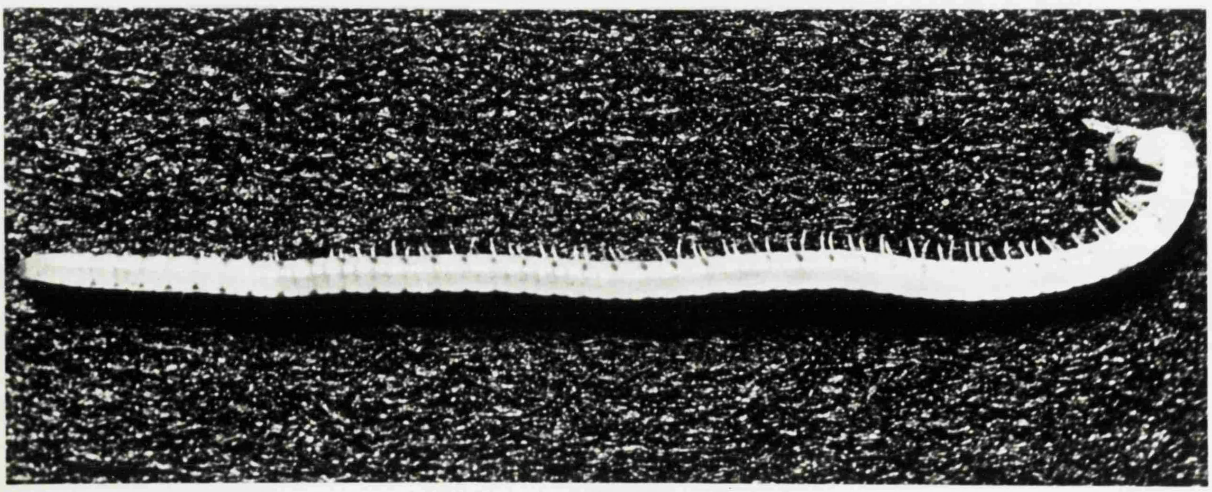
PLATE 19. STADIUM I BLANIULUS GUTTULATUS x 70

PLATE 20. SOME STADIA OF *BLANIULUS GUTTULATUS* SHOWING THEIR RELATIVE SIZES.

TOP:	STADIUM	II x 12.
MIDDLE:	"	VII x 12.
LOWER:	"	X or XI x 12.







6.2.2.2. CULTURE OF BLANIULUS GUTTULATUS

50 adult millipedes, collected from the field in June, 1971 and kept previously at 4°C, were placed in each of two polythene boxes (7.1 x 4.7 x 1.8 in.) with airtight lids. Moistened sterilised peaty clay soil was added to a depth of 1 in. on 18 August. To one box only, a food supply of raw pre-germinated sugar-beet seed was added at intervals. Both boxes were kept in darkness in a heated laboratory where the temperature varied from 15°C to 22°C. Observations were carried out weekly and the appearance of young stadia noted. No attempt was made to disturb the soil unduly whilst counting the millipedes since previous observations have suggested that any soil disturbance may induce mortality particularly if the millipedes are moulting.

6.2.2.3. LIFE HISTORY OF SOME BLANIULIDAE IN SUGAR-BEET FIELDS

Information on the life history of blaniulids from the field, particularly their rate of development and appearance of different stadia, was obtained from regular collections from experimental sites at Bottisham (Cambs), Boston (Lincs), Magdalen (Norfolk), Shouldham (Norfolk), Kettering (Northants) and Marham (Norfolk) from 1971-1973; in addition, a field at Cambridge was sampled. Usually the first soil samples were taken, prior to sowing the crop in early spring, from varying depths but later samples were often from seed-spacing and herbicide experiments. At two sites, Shouldham and Cambridge, the soil was sampled again in November after harvesting. Cores were taken from either bare soil between the seedlings or from around the seedlings to a depth of between 3.9 and 13.4 in. Details of sampling at the different sites are shown in table 26.

After having extracted the millipedes from the soil by flotation using petroleum-spirit, the different stadia were separated under the microscope and numbers in each stadium category recorded: II, III-VI, and over VII (the adult stadia).

6.2.2.4. VERTICAL DISTRIBUTION OF BOREOIULUS TENUIS IN THE SOIL

At Shouldham 3.9 in. strata to a total depth of 13.4 in. were ~~was~~ sampled from April until November 1972 using a 2.3 x 3.9 in. auger; usually 10 cores were taken at random on bare soil or between the plants from each stratum on plots which had no insecticide treatment but on 20 April 24 cores were taken from two 3.9 in. deep strata to a depth of only 8.7 in. No soil samples were taken from July until November due to the hardness of the soil. The millipedes were extracted by flotation using petroleum-spirit.

TABLE 26 SAMPLING BLANIULID POPULATIONS FOR SEPARATION OF STADIA

Site	Sampling date	Maximum sampling depth (in.)
Bottisham	22.2.71	6
	14.4.71	8.7
	4.6.71	3.9
Boston	25.2.71	6
Cambridge	15.11.71	3.9
Magdalen	16.3.72	13.4
Shouldham	28.3.72	13.4
	6.4.72	"
	20.4.72	8.7
	23.5.72	"
	20.7.72	"
	13.11.72	"
Kettering	25.4.73	3.9
	14.5.73	"
	30.5.73	"
Marham	12.4.73	3.9
	8.5.73	"
	17.5.73	"
	6.6.73	"

6.2.3. RESULTS

6.2.3.1. CULTURE OF BLANIULUS GUTTULATUS

Several second stadia were observed in both boxes in the laboratory on 25 November approximately three months after the introduction of the adults. The original stock of millipedes were kept at 3-4^{o}C since at this temperature they lived longest but they failed to reproduce. By the end of January 1972 all millipedes in the box to which seeds had not been added had died but two stadium II and two stadium IV were observed in the other box. A single stadium V was seen in the soil on 23 March. On 4 April two stadium II and two stadium III were observed feeding around the embryo of a sugar-beet seedling within the fruit coat. One stadium I and one stadium VI was observed on 8 November; the former was inactive beneath the soil surface and is shown in plate 19. Some other stadia are depicted in plate 20 which shows their comparative sizes.

6.2.3.2. LIFE HISTORY OF SOME BLANIULIDAE IN SUGAR-BEET FIELDS

Fig. 27 shows the percentage of blaniulids in each of the three age categories at Bottisham in 1971, Shouldham in 1972, Kettering and Marham in 1973. In samples taken until June at three sites and November at one site B.tenuis was the millipede most commonly found in large numbers. B.guttulatus occurred in small numbers at Boston where 3 stadium II were found on 25th

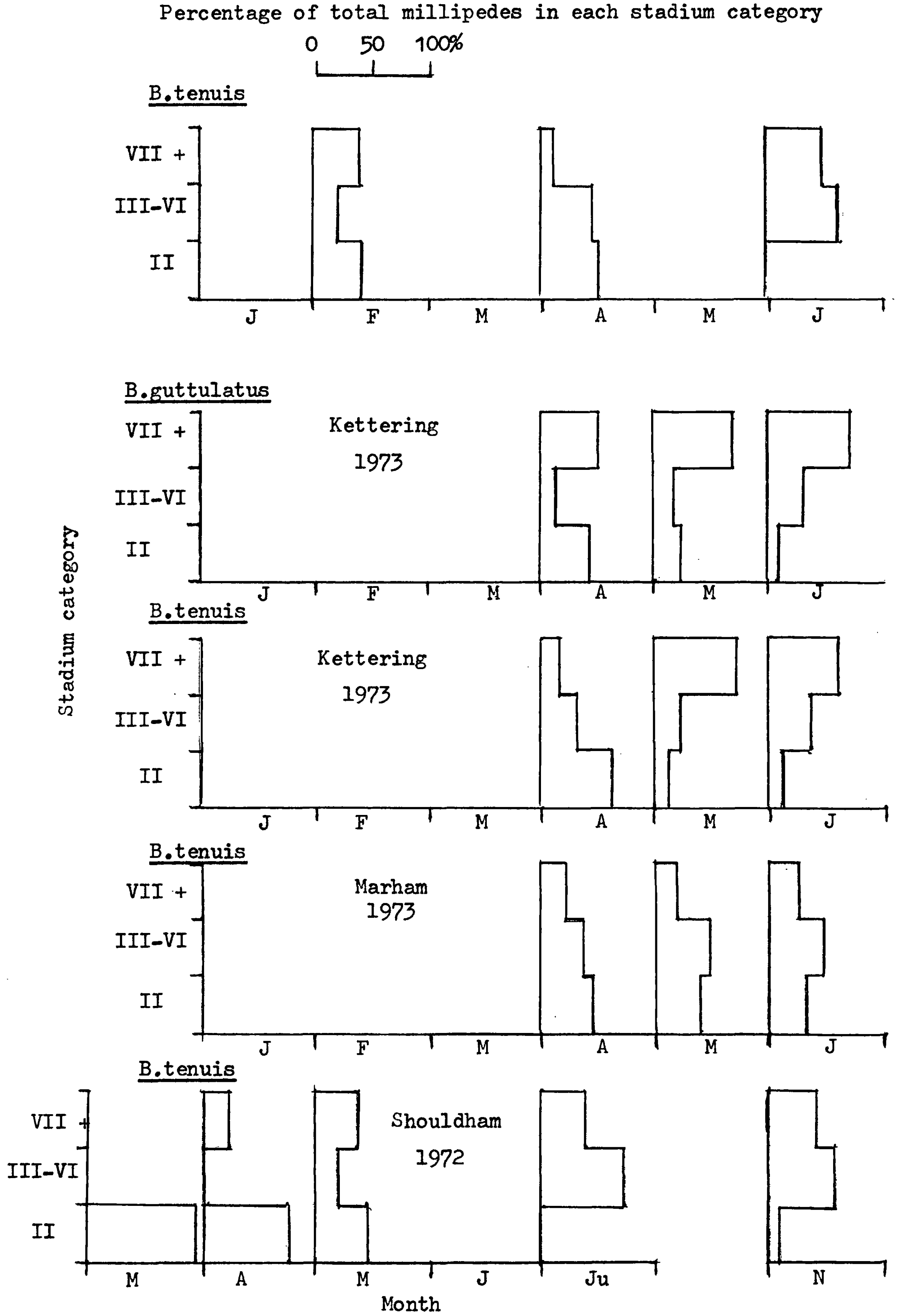


FIG. 27. AGE-STRUCTURE OF BLANIULIDS IN SUGAR-BEET FIELDS.

February and in a sample at Magdalen on 16 March where of the eight individuals extracted, three were stadium II, one stadium III and sections of four dead adults.

It was common to find mostly stadium II in samples taken in April, although at Marham two stadium I were recovered on 12 April but they comprised only 1% of the total. Stadium II were particularly numerous at Shouldham, where in March they formed 97% of the total blaniulids recovered and were still the most common stadium in May. At all sites the proportion of the total blaniulids in stadium II decreased in May and June samples whilst there was a corresponding increase in numbers of the other stadia. By July adults were sometimes the most numerous in soil samples and stadium II the least numerous. At Shouldham, where the field was sampled again in November 1972, 33% of the 33 millipedes recovered were stadium IV, no III were found but a few II; also at Cambridge in 1971 stadia II and III were found in samples taken in November. A small number of soil cores taken in April at Shouldham contained a mixture of pig-manure and cereal straw which had been applied to the soil and not thoroughly mixed. On 6 April 25% of the cores included this mixture and they contained 51% of the total stadium II in the samples. Also on 20 April 13% of similar cores contained 52% of all stadium II.

6.2.3.3. VERTICAL DISTRIBUTION OF BOREOIULUS TENUIS IN THE SOIL

Fig 28 shows the numbers of B.tenuis in each of the three strata in the soil from April until the end of July. Although a

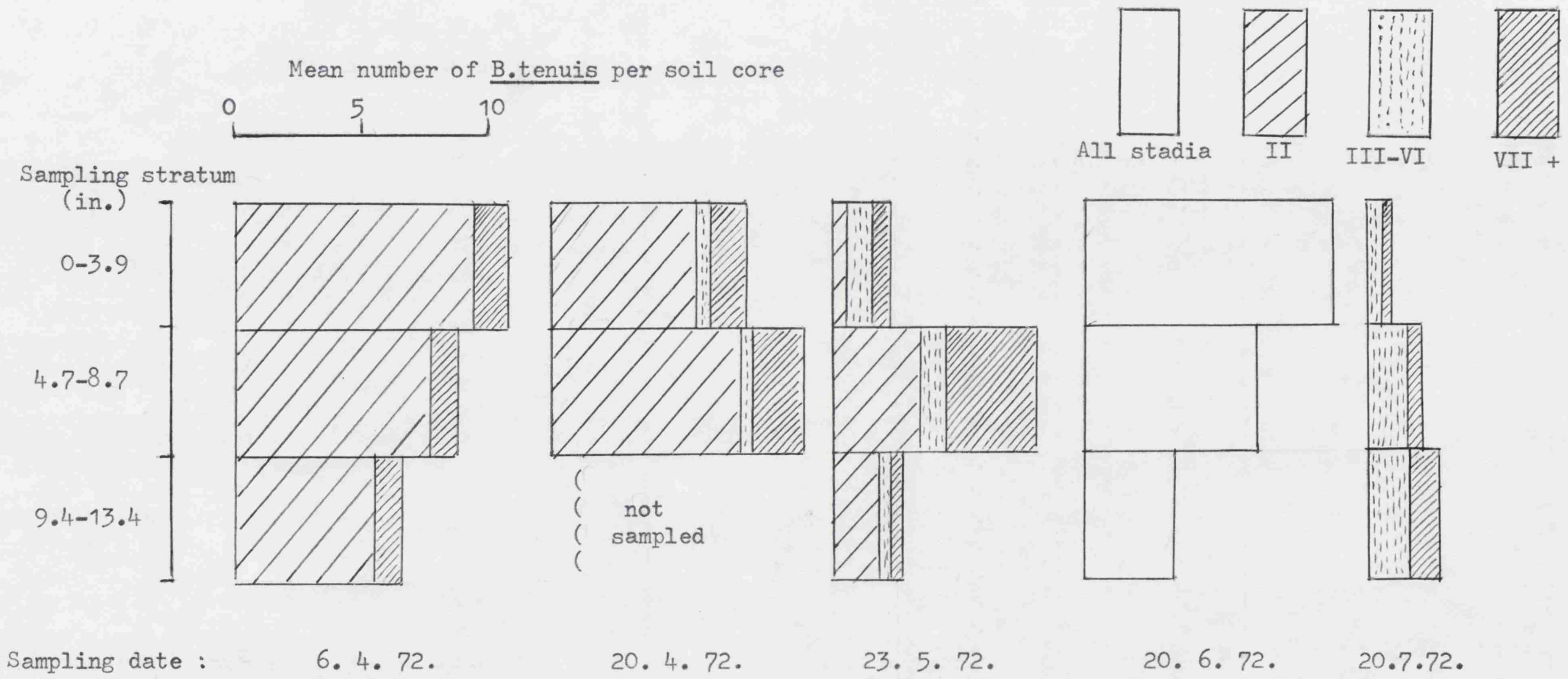


FIG. 28. VERTICAL DISTRIBUTION OF *BOREOIULUS TENUIS* STADIA IN THE SOIL AT SHOULDHAM IN 1972.

total of 10.8 per core were found in the top stratum in early April adults averaged only 1.2 per core and no intermediate stadia were found. In samples taken on 20 April adults in the top soil reached a peak with 1.8 and 2.2 per core in the 0-3.9 in. and 4, 7-8.7 in. strata respectively. Total numbers of millipedes declined in the top stratum in May, particularly stadium II, but adults averaged 3.5 per core in the middle stratum. Stadia were not separated in the June samples where there were 9.9 B.tenuis per core from the topmost sample. In July no stadium II were recovered from any stratum and most millipedes were present as III; for the first time most millipedes were found in the deepest samples but the total number of millipedes recovered was few.

6.2.4. DISCUSSION

The culture of B.guttulatus was unsuccessful; the high mortality of individuals did not allow a full cycle from adult through egg to adult to be completed and the culture was abandoned. The mortality was probably induced by the high temperature at which they were kept relative to that in their natural habitat, since in stocks of millipedes kept at $4^{o}C$ mortality was low. A high temperature was necessary to shorten the interval between moults and stimulate development and many dead millipedes were found in moulting chambers constructed in the box at the soil/plastic interface, particularly on the bottom. Kinkel (1955) reared B.guttulatus at 20-$25^{o}C$ and fed them partially decayed vegetables (potatoes, cucumbers, lettuce and germinating seed) as well as some animal material (earthworms, slugs and Tenebrio larvae). A suitable food material (sugar-beet seed) appeared to be necessary for millipede survival in the present study. Breny (1964) believed that egg production could not take place without the prior ingestion of living plant material and although first stadia were found in the breeding box to which fresh plant material had not been added they did not survive. Suitable food material may therefore be necessary as a substrate for the continuing development of the youngest stadia. It is perhaps significant that large numbers of second stadia were found in association with the manure and cereal straw mixture in the field at Shouldham but it is not known whether this medium provided a suitable spot for oviposition or whether the stadium II individuals had collected on it later. Absence of stadium I from this substrate is not necessarily evidence to lend weight to the migration theory; sampling may have been too late to record this stage. Since many eggs are laid at the same time, hatching and larval development would be largely synchronous thus effectively disguising preceding events. Stadium I are relatively immobile and remain in this stage for only 14 days (Breny and Biernaux, 1966). These workers also concluded that since they were to be found in the soil 20-40 cm. deep then egg-laying probably occurs at this depth. In the present study two stadium I were found in the top 3.9 in. (at Kettering in 1973) which would indicate that egg-laying can probably occur at any depth providing that conditions are suitable. There is therefore no evidence which contradicts the premise that the manure-straw mixture probably served as the location of oviposition and culture medium for the eggs and young stadia.

The presence of stadium II in April and again in November would indicate two breeding periods in the field as Kinkel (1955) had observed. His adult females, collected in the autumn, laid eggs after three weeks in the laboratory and he found that under the constant temperature and humidity conditions breeding periods were less pronounced than under field conditions. One can use the life-history data provided by this worker to interpret the course of events in the breeding boxes in the present study. If one allows eight weeks from oviposition to appearance of stadium II then the adult B.guttulatus introduced into the breeding boxes would have oviposited after about 4 weeks. The appearance of a stadium V individual four months after observing some second stadia agrees well with the development rate recorded by this worker of 4-4½ months for the equivalent development, or a total of 6 months from oviposition. The appearance of stadium II from November until the following April in the breeding box suggests a long oviposition period under suitable conditions. Kinkel also observed that eggs of B.guttulatus are particularly sensitive to changes in humidity and the failure to obtain young stadia from previous cultures, which were often disturbed, may be attributed to this sensitivity in the egg stage.

In the field, present observations indicate that stadium II, present from March until May in the field, have attained stadia IV to VI by the following November and hence the new generation would overwinter in these stadia. Breny and Biernaux (1966) calculated that if eggs were laid in April-May then the millipedes would have attained stadium V by the end of October. These workers also pointed to the difficulty of studying the development of B.guttulatus from month to month in the field since millipedes of all ages could be found in the soil at all levels. They calculated that B.guttulatus may reach stadium XVI or XVII and, allowing three moults per year, would result in a life span of 5 or 6 years.

In Belgium both A.pallidus and B.guttulatus show vertical displacements in the soil which can be correlated to changes in temperature and moisture at different depths, (Biernaux 1968; Biernaux and Baurant 1964a, 1964b; Pierrard, Boute and Baurant 1963). That the numerical superiority of stadium II compared with other stadia in Spring was not attributable to the shallow sampling was confirmed by the deeper sampling at Shouldham where the proportion of the total in this stadium was approximately constant at the three sampling depths in early April. However, at other sites such as at Kettering, where samples were taken from only the top 3.9 in. of soil, the rapid increase in the proportion of adults to other stadia in May cannot be adequately explained by the rate of maturation of the young stadia. The conclusion must be that the adults are the most mobile group and capable of moving up and down the soil profile more than the younger stadia. A shallow sample taken in Spring may not, therefore, estimate accurately the total numbers present in the soil or the age-structure of the population if others are present deep in the soil. Since the soil at the Kettering site had a good structure and depth, there was no apparent barrier to their movements but at Shouldham the soil was penetrable with the auger to a depth of only 13.4 in. below which was a layer of solid chalk; this may have served to restrict the penetration of the millipedes to within the shallow sampling zone. The low overall number recovered from the July sample would indicate either a high mortality or that many were present outside the sampling zone. Since the soil samples were taken between plants, some of which had been infested by large numbers of millipedes in the root-zone in June, many millipedes still remaining on the deeper portions of the root system may represent a significant proportion present in the soil and they would not be included. A further explanation would be that the millipedes had moved vertically out of the sampling zone into fissures in the underlying chalk, either in response to temperature or moisture changes, or for moulting. No moulting blaniulids, which assume a non-active state and become coiled prior to casting their skins, have ever been recovered in soil samples. This may be due to their tendency, observed in the laboratory, to move to the deepest layers in the soil to moult upon some firm surface or that the extraction method was inefficient for this stage. Also, laboratory observations at 7°C indicated that only a small proportion, and never more than 24% of the total available, fed on seedlings at any one time.

6.3. DISTRIBUTION OF MILLIPEDES IN ARABLE FIELDS

6.3.1. INTRODUCTION

Observations in sugar-beet crops in September and October have indicated that millipedes come to the soil surface and feed under fallen sugar-beet leaves lying on the soil surface. It is a time when pairing of Brachydesmus occurs and the adults, which form a high proportion of the population in autumn, can easily be found. Blaniulid millipedes, though less ubiquitous, also move near to the soil surface and may be found in similar habitats, sometimes together with B.superus.

A survey was conducted with the co-operation of the British Sugar Corporation in the Autumn of both 1970 and 1971. There are seventeen sugar factories operated by the Corporation; most are located in eastern England where arable farming is most intensive (Coppock, 1964). Sugar beet is grown in rotation under contract to each factory, but the acreage assigned to each factory varies from 10,000 to 40,000 acres. Responsibility for the sugar beet within each factory area is shared amongst a number of 'fieldmen'. The object of the survey was to assess whether millipedes are evenly distributed throughout all arable fields, to determine the most common species and record the soil type on which they are found. Most previous studies, on ecology and distribution, have been limited to woodland habitats (Blower, 1970; Blower and Gabbut, 1964) or dune systems (Barlow, 1958) and only scant attention has been given to arable habitats in England, particularly those receiving regular cultivation.

6.3.2. METHODS

In October of 1970 and 1971 a questionnaire to each factory fieldman at all factories requested their co-operation in searching sugar-beet fields for millipedes. Each fieldman chose four fields at random and searched in three separate square foot sampling points per field, each sampling point being well separated from the others. Any millipedes found were placed in a polythene bag together with some moist soil and a senescent sugar-beet leaf, on which the millipedes could feed. Each fieldman recorded location of the field, the date the sample was taken and the soil type together with the number of millipedes found in each of the three sampling points per field. In the laboratory the millipedes were identified and the numbers of each stadium of *B.superus* counted so that the age-structure of their populations could be compared in the two years.

6.3.3. RESULTS

6.3.3.1. Occurrence of different species

Table 27 shows the percentage contribution of each of the different species found in both years. B.superus was by far the most common millipede; it was found in 26 samples in 1970 and 30 samples in 1971 each from separate fields. Since the samples were taken at approximately the same time each year the age-structure can be compared. Samples in 1970 and 1971 comprised 296 and 285 individuals respectively and the percentage in each stadium were for each year respectively: VII, 55%, 47%; VI, 29%, 39%; V, 13%, 12%; IV, 3%, 3%. No younger stadia were found. The respective sex ratio for the adults was 2.1:1 in 1970 and 2.2:1 in 1971 in favour of the females. Whereas only 15 B.guttulatus were found in 1970 in only one field, a total of 63 were found in 6 fields in 1971. Of these all were adult except for 10 III and 2 II. Of the other six species of millipede recorded all were less common.

6.3.3.2. Comparison of numbers found in each sugar factory area

The location of each factory area is shown in plate 21. The relative abundance of millipedes (all species) in each sugar factory area was expressed as the number of millipedes found per fieldman per field (3 x 1 foot squares) separately for 1970 and 1971 (Table 28). Numbers varied for each survey; in some factory areas such as Peterborough and Selby, relatively high numbers were caught in both

TABLE 27 RELATIVE ABUNDANCE OF THE DIFFERENT MILLIPEDE SPECIES IN THE 1970 AND 1971 SURVEYS

Millipede species	Percentage of total millipedes in samples	
	1970	1971
Brachydesmus superus Latzel	89.9	77.0
Polydesmus angustus Latzel P.gallicus Latzel	5.1	0.6
Archeboreoiulus pallidus (Brade-Birks)	0	0.3
Blaniulus guttulatus (Bosc)	4.5	20.4
Brachyiulus pusillus (Leach)	0	1.0
Ophyiulus pusillus (Leach)	0.3	0.3
Cylindrioiulus spp.	0.3	0.3

PLATE 21. SUGAR FACTORY AREAS, LOCATION AND ACREAGE

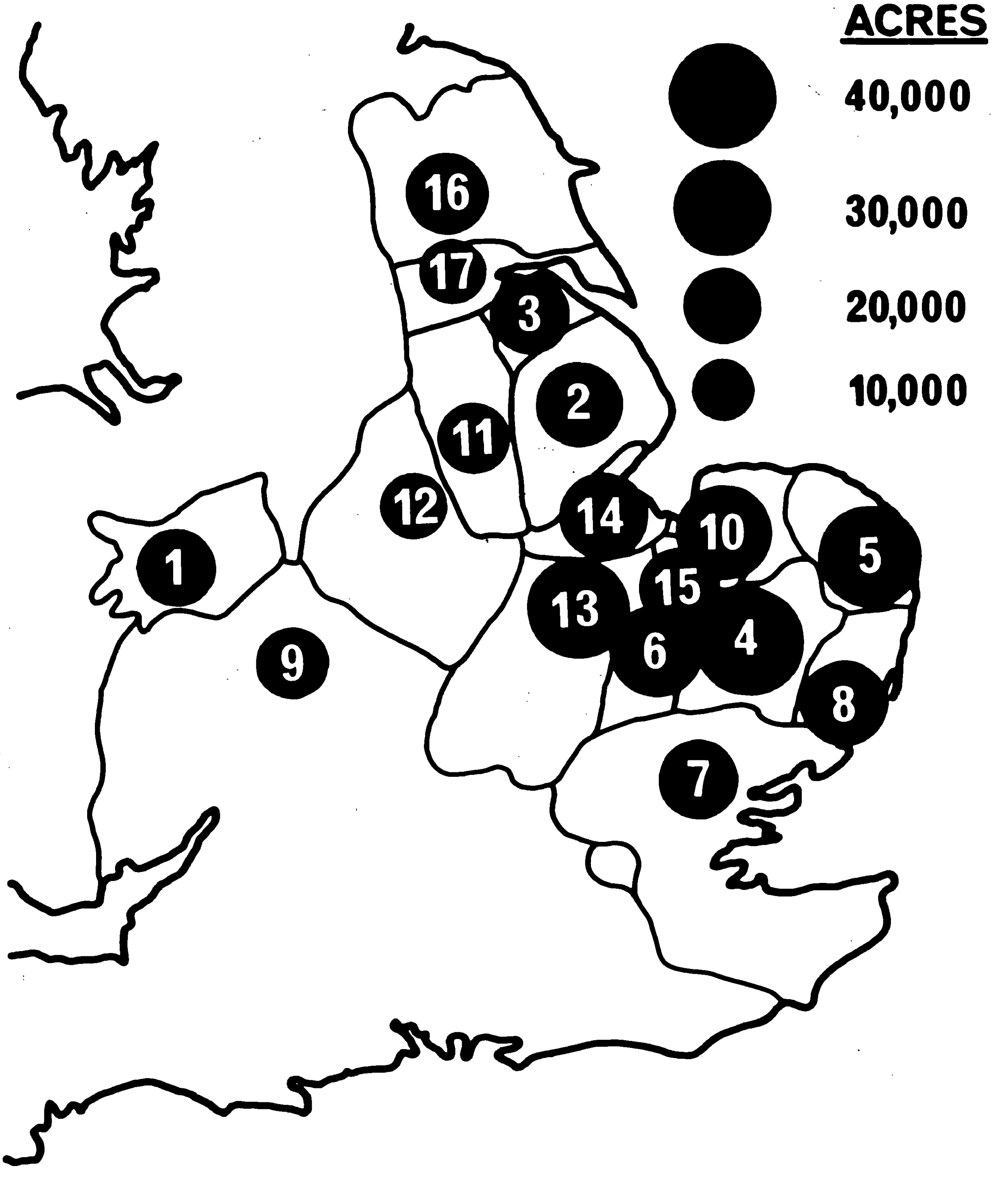

ACRES
40,000
30,000
20,000
10,000
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17

TABLE 28 THE RELATIVE ABUNDANCE OF MILLIPEDES IN EACH SUGAR FACTORY AREA

Numbers of millipedes found per fieldman per field

	Sugar factory area	1970	1971	Mean for 1970 & 1971
1	Allscott	0.1	3.4	1.8
2	Bardney	1.4	0.1	0.8
3	Brigg	0.5	0.1	0.3
4	Bury St. Edmunds	0	0	0
5	Cantley	0.2	0.5	0.4
6	Ely	2.6	0.4	1.5
7	Felsted	0	0	0
8	Ipswich	2.4	0.7	1.6
9	Kidderminster	0	1.8	0.9
10	King's Lynn	0.7	0	0.4
11	Newark	0.5	3.7	2.1
12	Nottingham	0	0	0
13	Peterborough	12.6	3.3	8.0
14	Selby	6.2	3.8	5.0
15	Spalding	1.8	2.5	2.2
16	Wissington	0.5	0.1	0.3
17	York	0.6	0	0.3

years wheareas in others, Bury St. Edmunds, Felsted and Nottingham, none were found in either year. Taking a mean for both years the factory areas can be ranked in decreasing order according to the number of millipedes found per fieldman per field: Peterborough 8.0; Selby 5.0; Spalding 2.2; Newark 2.1; Allscott 1.8; Ipswich 1.6; Ely 1.5; Kidderminster 0.9; Bardney 0.8; Cantley 0.4; Brigg, Wissington and York 0.3; Bury St. Edmunds, Felsted and Nottingham, 0.

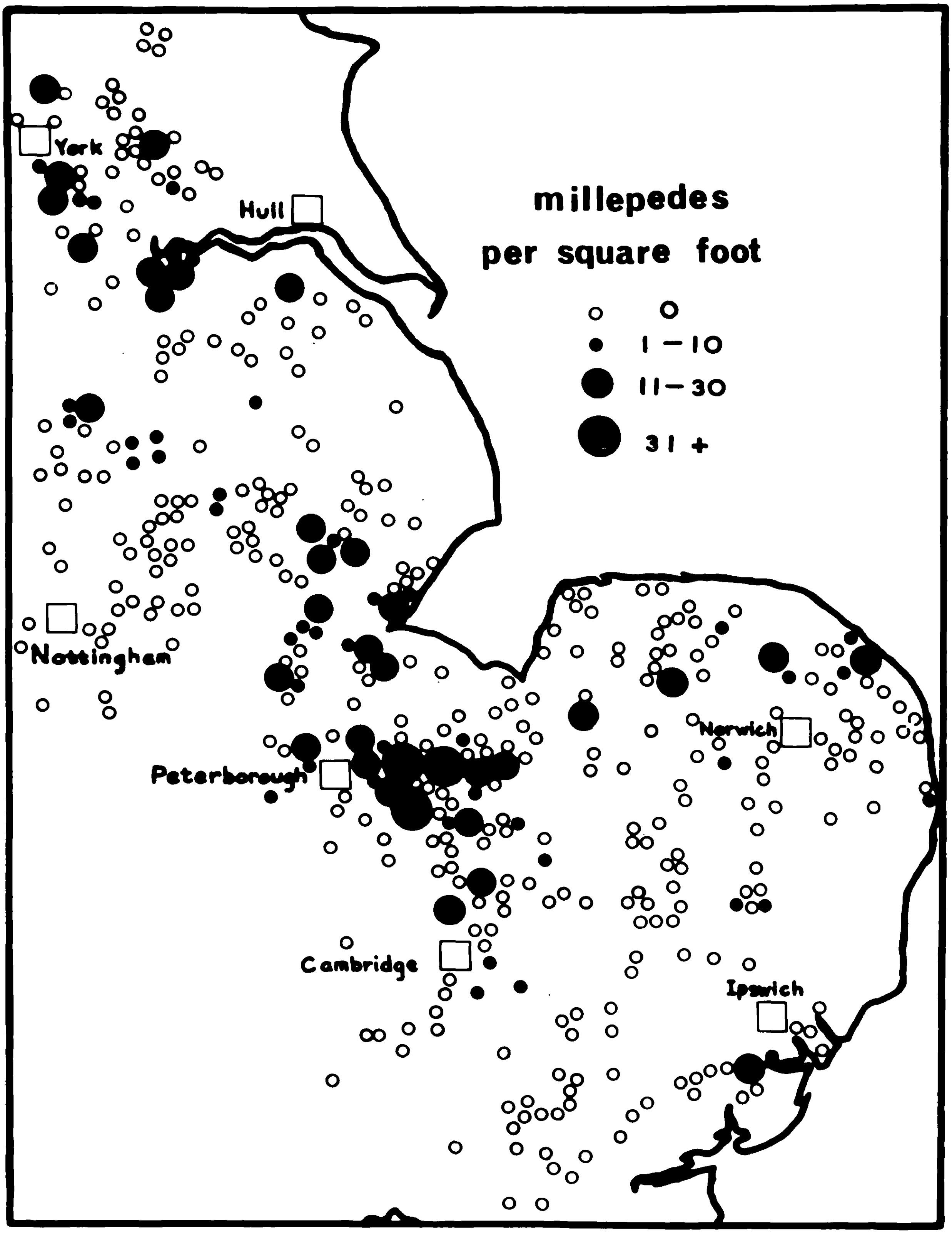
6.3.3.3. Geographical distribution of millipedes

Plate 22 shows the geographical distribution of the fields searched in the two surveys in eastern England, the most important sugar-beet growing area. The map does not show the fields in western England that were searched. Indicated are those fields where millipedes were either found or not found; the number of millipedes per square foot are shown as black circles of different radii. The absence of searches in some areas usually indicated a scarcity of arable fields but some reflected a lack of response of a fieldman to the questionnaire. Millipedes were found in 85 fields (15%) out of a total 578 different fields searched. The greatest frequency of finding millipedes was in the area immediately east of Peterborough on the Bedford level and in the area near Goole (Yorks.) but they were also found in scattered fields throughout Yorkshire, Lincolnshire, Nottinghamshire, Bedfordshire, Norfolk, Suffolk and Essex.

6.3.3.4. Millipedes on different soil types

Fig. 29 shows a comparison between the distribution of soil types on the total fields searched and those where millipedes were found. Data presented individually for 1970 and for 1971, from which mean values were calculated, is presented in appendix table 2. Most fields visited (26%) were sandy loams and most of the fields with millipedes (24%) were also of this soil type. Millipedes were found on most soils except loamy very fine sand. Generally their frequency of occurrence on the different soil types reflected the frequency of the soil types in the total fields

PLATE 22. GEOGRAPHICAL DISTRIBUTION OF MILLIPEDES IN EASTERN ENGLAND PLOTTED FROM MILLIPEDE SURVEY REPORTS, 1970 AND 1971.



MILLEPEDE DISTRIBUTION :

AUTUMN SURVEYS 1970 AND 1971

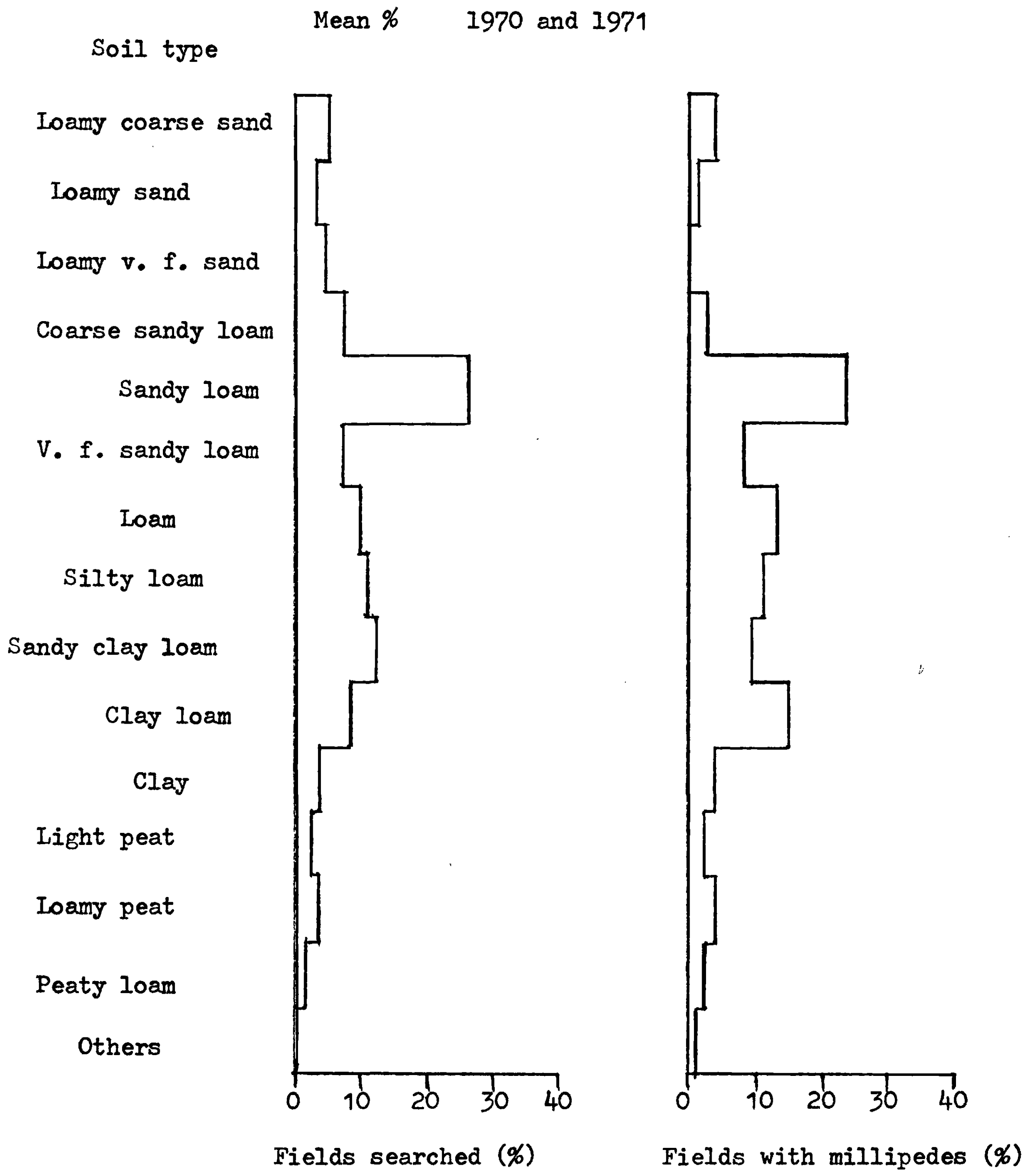


FIG. 29. A COMPARISON BETWEEN THE DISTRIBUTION OF SOIL TYPES ON THE TOTAL FIELDS SEARCHED AND THOSE WHERE MILLIPEDES WERE FOUND.

searched but there were some exceptions. Comparing the two histograms reveals two trends. Firstly, fewer fields of coarse soil texture, viz: loamy coarse sand, loamy sand, loamy very fine sand and coarse sandy loam appear to support millipede populations. Secondly, there was a trend for more fields of fine soil texture, viz: very fine sandy loam, loam and particularly clay loam to have millipedes.

6.3.4. DISCUSSION

Fieldmen are each responsible for 2000-5000 acres of sugar beet. In the 1970 survey the fields chosen by one fieldman were sometimes close to one another and were not representative of the potential area. In 1971 it was specifically recommended that fields should be at least 5 miles apart in order to improve the sample. Undoubtedly in an area where many soil type variations were present the number of sample points per field and number of fields sampled would be unrepresentative.

It would be difficult to improve upon the existing sampling method in practice since the fieldmen were untrained observers whose diligence varied. Since one of the objectives was a mapping of millipede distribution then a more precise technique would be one based on the 10 Km. National Grid square standard on British Ordnance Survey maps which is now being extended to record myriapod habitats (Barber and Fairhurst, 1974).

Since the survey was essentially one of sampling the surface soil in sugar-beet fields then there would be a bias in favour of the more surface-active species and hence it is not surprising that a polydesmid B.superus was most common. Blower (1970) found Polydesmus to be capable of and to indulge in much more surface activity than iulids. Almost all the fields recorded having millipedes included B.superus as the main species but it was also more widespread than any other species. The survey also favoured those species with an autumn activity peak such as B.guttulatus which inhabits the top soil layers when sufficiently moistened (Biernaux, 1968). In woodland most of the activity of Iulus and Tachypodiulus occurs in April, May and June and Polydesmus augustus and P.denticulatus are mainly active in mid-summer (Blower, 1970) but patterns of activity and their life-cycle may be different in arable habitats. In pitfall trapping for soil-surface inhabiting arthropods peak numbers of B.superus and Polydesmus gallicus were found in September 1970 and most individuals were seen under decaying sugar-beet leaves lying on the soil surface (Baker, 1971).

Most B.superus were adults and the sex ratio was approximately constant at 2 females to 1 male in both surveys; the age structure was also very similar. A few fields indicated high populations of B.guttulatus in the 1971 survey and in one of these the sugar-beet seedlings had suffered severe damage in the spring. As in other studies, some very young stadia were found in the soil from under the fallen beet leaves sent to the laboratory, indicating an autumn breeding period for this blaniulid millipede.

Both Peterborough and Selby factory areas stand out as having more millipedes in their fields than other areas. Many more were found at Newark in 1971 than 1970, half were B.guttulatus in 1971 when it became more prominent. Since this species is the one most often associated with damaged sugar-beet seedlings then the apparent increase in this species in 1971 may provide a partial explanation of increased incidence of damage by this species compared with 1970 (Dunning, 1975). Generally those factory areas where most millipedes were observed in the surveys, e.g. Peterborough, also had a record of the highest incidence of reported damage to sugar-beet seedlings in the spring by fieldmen (Dunning, 1975).

The geographical feature which can be fitted to most of the areas where millipedes were found are rivers and low-lying marshy ground, e.g. fenland bordering the western edge of the Wash and marshland near Goole on the river Humber. Kinkel (1955) found that the distribution of B.guttulatus in Germany tended to follow the pattern of human settlements particularly in river valleys where the forest had been cleared to make way for cultivations.

The distribution of soil types in the survey was a close reflection of the relative percentages of each type on a national scale (Rose, 1972). Since millipedes were found on all soil types no distinct preference could be discerned from the surveys. However, other workers have found millipedes to be more common on certain soil types but no accurate surveys have been reported. Edwards (1929), investigating the fauna of pasture and agricultural land, found B.guttulatus on only one of the five soil types sampled; in Belgium Breny and Biernaux (1966) reported that B.guttulatus and Archeboreoiulus pallidus are most common on silty clay arable soils and absent from well drained sandy soils. It may well be that the soil's moisture holding capacity is the most important feature and a range of soils may be of suitable type.

Other factors which may affect the distribution of millipedes such as soil organic content and use of farmyard manure were not investigated in the surveys. Large differences in sizes of populations of Diplopoda on dunged and undunged plots were reported by Morris (1927); the millipedes, mainly B.guttulatus, averaged 1.7 million on the dunged plots compared with only 0.9m. on plots not receiving dung. In the present studies at experimental sites large numbers have been recorded on ploughed-in straw and FYM, decayed sugar beet and pea haulm but these materials may serve only to retain the millipedes near the soil surface or in the top soil where application was concentrated.

7. SOME ASPECTS OF THE BEHAVIOUR OF MILLIPEDES

7.1 REACTIONS TO SOIL MOISTURE

7.1.1 Introduction

Moisture is present in the soil both as a film of free water surrounding the soil particles and in a gaseous state in the soil atmosphere. Comparatively large amounts of water have to be removed from soil to lower the equilibrium humidity in the soil atmosphere below its saturated state (Puri, Crowther & Keen, 1925). Variations in humidity in the soil will therefore be rare under most conditions and likely only to affect the topmost layers. Differences in Relative Humidity have been found to affect the distribution of millipedes such as Proteroiulus fuscus (am Stein), which leave the soil to inhabit old tree stumps (Peitsalmi, 1974) and Oxidus gracilis (C.L.Koch) (Perttunen 1953); however, within the soil the amount of free water around the soil particles is likely to show more variation than Relative Humidity. Observations in arable fields in Belgium have suggested that soil moisture is a factor governing the seasonal activity and vertical distribution of millipedes in the soil (Biernaux, 1968).

In the present study the effect of different soil moisture régimes on both the horizontal and vertical distribution of B.superus and B.guttulatus was studied in the laboratory under constant humidity conditions. The possibility of a geotactic response was investigated by recording the vertical movements of B. guttulatus in soil of uniform moisture content. The influence of germinated sugar-beet seed on their vertical movements in the soil was also studied.

7.1.2 Materials and Methods

All the experiments were conducted in boxes of soil, 8.0 x 1.7 x 1.7 in., constructed of 'Perspex' and fitted with detachable lids. Soils used were of various moisture contents according to the particular experiment but all were of the same crumb size. The soil, a very fine sandy loam, collected from the millipedes natural habitat was shaken and passed through a 0.1 in. sieve to produce uniformly sized crumbs which, when packed together, would allow interstitial penetration by the millipedes. In later experiments the soil was oven-dried to constant weight and soils of different moisture content prepared.

7.1.2.1 Effect of soil moisture on the distribution of Brachydesmus superus.

In this experiment four treatments were used with boxes of soil; A 'moist' soil, was collected from the field from around the base of sugar-beet plants where many B.superus could be found; B 'dry' soil, was from between the rows where there were no millipedes; C 'half-dry' soil, 'half-moist' soil - millipedes in the moist side and D 'half-dry, 'half moist' - millipedes released in the dry side. In filling boxes C and D a cardboard partition was made to keep the soils in each half separate until the boxes were completely full, after which it was withdrawn. Lids were replaced, held in position by tight string bands and the boxes stored in the dark at laboratory ambient temperature (15 - 22°C) for an initial period of 4 hrs. for humidity to become uniform throughout. Lids were then partially removed and thirty adult B. superus were released at one end (marked X). After 24 hrs. the soil in each half of the boxes was tipped separately onto a sheet of paper and the number of millipedes in each counted. The experiment was repeated for nine consecutive 24 hr. periods (trials) using fresh millipedes and freshly prepared soil at each trial. The objectives were to assess the amount of movement in both soil of uniform moisture content and movement towards a soil providing an alternative moisture régime.

7.1.2.2. Effect of soil moisture on the distribution of Blaniulus guttulatus.

A sample of soil (very fine sandy loam) collected from the field, was oven dried to constant weight. The soil was spread thinly and divided into four on metal trays. Moisture contents of 10, 15, 20 and 29% were obtained by wetting the soil with a fine, evenly dispersed spray of a calculated volume of water derived from the relationship:

$$\text{m.c.} = \frac{(A + W) - A}{A + W} \times 100\%$$

where A = wt. of oven-dry soil

W = wt. of moisture in the soil

m.c. = moisture content of soil

Gradients of increasing soil moisture were established in four Perspex boxes by filling successive sections (separated by cardboard partitions while filling) with soils of increasing m.c. prepared according to the method outlined above. The boxes were fitted with air-tight lids held with string bands and left for approximately 4 hrs. in the laboratory at 15 - 20°C for the humidity in each box to reach a maximum. This was marked by the appearance of condensation on the interior faces of the plastic. Lids were partially removed briefly to allow the introduction of 20 stadium VI B. superus to sectors with soil of 29% m.c. in two boxes and sectors of 10% m.c. in the remaining two boxes. In the first series of experiments the boxes were stood vertically with millipedes in the top sector in two boxes and the bottom sector in the other two boxes. At 24 hr. intervals the soil was removed and the number of millipedes in each sector counted. The experiment was repeated four times (trials), after which the boxes were used horizontally and the millipedes placed into the sector containing the driest soil (10% m.c.). The experiment was repeated six times (trials) using fresh millipedes and soil in each trial. The objectives were to assess the millipedes soil moisture preferences and investigate any response to gravity by comparing the direction of movement through a soil moisture gradient in upward and downward directions.

7.1.2.3 The influence of germinated sugar-beet seed on the vertical movements of millipedes.

Uniformly moist peaty loam soil with a moisture content of approximately 29% and a crumb size of not more than 0.1 in. was packed loosely into four 'Perspex' boxes (8.0 x 1.7 x 1.7 in.). The boxes were left for 4 hrs. at 15-20^{o}C for the humidity to become uniform throughout. Five raw sugar-beet seed which had been left to germinate for 3 days on moist filter paper in the laboratory were placed in the top of one box and the bottom of another box which stood vertically. Between 11 and 15 B. guttulatus were released at the introduction point (marked X) at different ends of each of the four boxes so that their direction of net vertical movement within bare soil could be monitored and compared with boxes where the top or bottom ends contained germinated sugar-beet seed. The boxes were kept in darkness in the laboratory at 15-20°C. At 24 hr. intervals the soil in each of the boxes was removed, the millipede distribution in the boxes was plotted by subdividing them into 4 equal sectors 2 in. long, as in previous experiments, using cardboard partitions. The numbers of millipedes in each sector per box was counted. The experiment was repeated six times (trials) using fresh millipedes and soil.

7.1.3. Results

7.1.3.1 Effect of soil moisture on the distribution of Brachydesmus superus

Table 29 shows the comparative net millipede movement both in soil of uniform moisture content and where an alternative moisture was available within the same box. The numbers of millipedes in each half is expressed as a percentage of the total number originally introduced into the half of the box marked X.

In uniformly 'dry' soil (13% moisture content) 59% of individuals had moved away from the point of introduction, X, compared with 38% in uniformly 'moist' soil (21% moisture content). Where an alternative moisture was provided 70% moved out of 'dry' soil into 'moist' soil but only 16% had moved in the opposite direction.

7.1.3.2 Effect of soil moisture on the distribution of Blaniulus guttulatus

A convenient scoring system was adopted to compare the amount of millipede movement in each of the boxes. It was based on the net distance moved by one millipede from the introduction point, X; 24 hr, recordings gave the number of millipedes at rest in each of the four sectors in the boxes and from this data a standard measure of the net distance travelled was calculated according to the following convention for each millipede :

TABLE 29 RECORDINGS OF THE MOVEMENT OF *BRACHYDESMUS SUPERUS* IN SOIL OF BOTH UNIFORM MOISTURE CONTENT AND WITH AN ALTERNATIVE MOISTURE LEVEL

Percentage of *B.superus* in each of two halves of the boxes after 24 hrs. (X = release point)

	Soil of uniform moisture				Soils of alternative moisture			
	Box A		Box B		Box C		Box D	
Trial	'Moist'	'Moist'	'Dry'	'Dry'	'Moist'	'Dry'	'Dry'	'Moist'
1	80x	20	30x	70	57x	43	43x	57
2	57x	43	77x	23	70x	30	47x	53
3	60x	40	17x	83	93x	7	47x	53
4	50x	50	37x	63	70x	30	27x	73
5	67x	33	77x	23	90x	10	23x	77
6	67x	33	3x	97	90x	10	3x	97
7	67x	33	27x	73	100x	0	7x	93
8	63x	37	30x	70	100x	0	40x	60
9	43x	57	73x	27	85x	15	30x	70
Mean %	62x	38	41x	59	84x	16	30x	70

Score 0 = No movement from point X. All millipedes in sector 1

" 1 = Movement of one millipede to sector 2

" 2 = " " " " " " 3

" 3 = " " " " " " 4

An Activity Index I was calculated for the millipede movement in each box and since sometimes different numbers of millipedes were used in the experiment, the Index was based on a score for a standard number of 100 millipedes. Table 30 shows the effect of soil moisture gradients on the vertical distribution and activity of B. guttulatus. When they were initially placed in the sector having soil of 10% m.c. 81% of the millipedes moved downwards into soil of the greatest available moisture but only 44% moved to this sector, in the opposite end of the box, in an upward direction. When placed in soil of highest moisture content 37% remained when released in top sector but 69% remained when they were released in the lowest sector of the box. Activity was always greatest for millipedes moving out of soil of lowest moisture content. For downward movement from both soil of high or low moisture the total Activity Index score was 389 compared with 214 for total upward movement.

Table 31 shows how B. guttulatus becomes distributed in a horizontal soil moisture gradient in 4 trials where the millipedes were released in sector X with a soil moisture of 10%. A mean of 50% reached sector 4 with 29% moisture after 24 hrs. A few died at the release point in Sector 1 but these were included in the calculations; the 17% in this sector is therefore an over-estimate.

TABLE 30. VERTICAL DISTRIBUTION & ACTIVITY OF BLANIULUS GUTTULATUS IN A SOIL MOISTURE GRADIENT.

Percentage B.guttulatus in each sector after 24 hrs. after release in sector marked X

Sector	m.c. %	Trial 1	Trial 2	Trial 3	Trial 4	Mean
1X	10	0	0	6	10	4
2	15	0	0	0	20	5
3	20	0	16	12	10	10
4	29	100	84	82	60	81
Activity Index		300	284	272	224	270
Sector	**m.c. %**	**Trial 1**	**2**	**3**	**4**	**Mean**
1X	29	22	44	46	35	37
2	20	28	33	23	10	24
3	15	22	17	30	23	
4	10	28	6	8	25	16
Activity Index		156	78	95	145	119
Sector	**m.c.%**	**Trial 1**	**2**	**3**	**4**	**Mean**
4	29	30	20	50	48	44
3	20	30	33	5	38	32
2	15	10	20	20	10	19
1X	10	30	27	25	10	26
Activity Index		160	110	180	229	170
Sector	**m.c.%**	**Trial 1**	**2**	**3**	**4**	**Mean**
1	10	0	15	0	0	4
2	15	5	5	9	5	6
3	20	19	20	27	19	21
4X	29	76	60	64	76	69
Activity Index		29	75	42	29	44

TABLE 31 DISTRIBUTION OF BLANIULUS GUTTULATUS IN A HORIZONTAL SOIL MOISTURE GRADIENT

Percentage B.guttulatus in each sector after 24 hrs. after release in sector marked X.

Sector	1X	2	3	4	Activity Index
m.c.%	10	15	20	29	
Trial 1	15	10	25	50	210
2	28	11	28	33	167
3	20	10	10	60	210
4	21	11	26	42	189
5	15	5	15	65	230
6	5	20	25	50	220
Mean	17	11	22	50	204

7.1.3.3 The influence of germinated sugar-beet seed on the vertical movements of millipedes.

Activity Indices, based on the activity of 100 millipedes, were calculated as in the previous experiment and the percentage of millipedes in each sector were calculated for both top and bottom release points and where germinated sugar-beet seed were included in the soil (Table 32). When B.guttulatus was released in the uppermost sector of the box 48% reached the bottom in 24 hrs. and 19% remained in the top. The inclusion of germinated sugar-beet seed at the top of the box increased the percentage remaining in the top sector to 65%; activity was also correspondingly less. When the millipedes were released into the bottom sector of the box 10% were found in the top sector after 24 hrs. compared with 50% when seeds were placed at the top of the box; the seed in both boxes appeared to cause a redistribution of the millipedes. Comparing the two 'control' boxes without seed, net downward movement was greater than movement upwards in the boxes as in previous experiments, even though no moisture gradient was present (mean Activity Indices of 201 versus 84).

TABLE 32. EFFECT OF GERMINATED SUGAR-BEET SEED ON THE DOWNWARD MOVEMENT AND DISTRIBUTION OF *BLANIULUS GUTTULATUS* IN UNIFORMLY MOIST SOIL COLUMNS.

1. Control

Percentage *B.guttulatus* in each sector after 24 hrs. after release from sector marked X.

Sector	m.c.%	Trial 1	2	3	4	5	6	Mean
1X	29	20	43	14	0	27	7	19
2	29	7	14	21	9	0	8	10
3	29	47	7	36	18	6	31	24
4	29	26	36	29	73	67	54	48
Activity Index		180	136	179	264	213	231	201

2. Germinated sugar-beet seed in Sector 1

Sector	m.c.%	Trial 1	2	3	4	5	6	Mean
1X	29	73	69	83	8	80	79	65
2	29	7	8	9	42	9	7	12
3	29	7	8	0	0	0	0	3
4	29	13	15	8	50	20	14	20
Activity Index		60	69	33	191	60	50	77

continued on P. 193

TABLE 32 (CONTINUED)

1. Control

Percentage B.guttulatus in each sector after 24 hrs. after release from sector marked X.

Sector	m.c.%	Trial 1	2	3	4	5	6	Mean
4	29	13	0	0	20	0	27	10
3	29	13	31	0	6	21	13	14
2	29	7	31	50	7	43	13	25
1X	29	67	38	50	67	36	47	51
Activity Index		73	92	50	80	86	120	84

2. Germinated sugar-beet seed in sector 4

Sector	m.c.%	Trial 1	2	3	4	5	6	Mean
4	29	50	43	67	25	67	50	50
3	29	21	15	11	25	8	25	18
2	29	8	21	11	0	8	17	11
1X	29	21	21	11	50	16	8	21
Activity Index		200	179	233	125	225	217	197

DISCUSSION

Millipedes are always found in parts of the soil profile that offer sufficient moisture. Biernaux (1968) found most *Blaniulus guttulatus* in soil where the moisture content was approximately 15%. fewer ~~seldom~~ where it was less than 11% and few at 20%. In the present study both species showed a sensitivity to moisture and a consistent preference in the horizontal boxes for soil of 29% moisture content compared with the other, less moist, soils. *B.superus* showed greater activity in 'dry' soil conditions in the experiments and both species moved less when initially introduced into moist soil. This is probably an orthokinetic response, i.e. a simple effect on the rate of locomotion which depends on the intensity of stimulation (Frankel & Gunn, 1940). Cloudsley-Thompson (1950) found that the millipede *Paradesmus gracilis* (Koch) as well as two woodlouse species, were stimulated into activity when the damp filter paper, upon which they had been resting, became dry. However, he concluded that for *Paradesmus*, orthokinesis, induced by dry soil conditions, does not depend on the physiological state of the animals water relations which had been suggested by Scharmer (1935) for a *Lithobius* sp. centipede. When placed in soil in which it is free to move in a vertical plane, *Blaniulus* moves both downwards and upwards from soil of low to high moisture where it comes to rest. Dry soil appeared to be a greater stimulus for downward rather than upward movement. Their vertical movements in a uniformly moist soil reinforced the previous indication of a positive response to gravity. However, Cloudsley-Thompson (1950) found that a positive geotaxis, evident in *Paradesmus* and to a lesser extent in *Blaniulus*, did not occurr in uniformly moist conditions but his observations were for periods of only two hours. The presence of germinated sugar-beet seedlings placed in the top-most sector of the vertical soil columns modified their normal positive geotaxis and appeared to attract millipedes to move upwards. It is not known whether the germinated seed merely served to arrest locomotion of previously wandering millipedes, which would otherwise have come to rest mainly near the bottom of the vertical soil column, or whether it was a positive attraction involving chemosensory perception. The concept of a positive geotaxis is at variance with the knowledge of their movements in the soil which involves mass upward movement in spring (Pierrard, Bonte & Baurant, 1963; Biernaux & Baurant, 1964b; Blower, 1970), but it is conceivable that it may occur at some time in their life cycle. It would be of definite survival value for a positive geotaxic response to occur prior to moulting and this can be observed in laboratory kept millipedes which move down deep into the soil of the culture medium to construct moulting chambers on some hard surface, usually the glass of their container. Experiments have not been conducted with Blaniulus collected fresh from the field when they are most numerous, in the spring; these individuals may not respond in the same manner.

7.2 EFFECT OF TEMPERATURE ON ACTIVITY

7.2.1 Introduction

Temperature fluctuations in soil are small, owing to its good insulating properties and ~~it~~ they will vary more in the topmost layers than deeper down, consequently the most responsive will be those arthropods which at some time can be found on or near the soil surface. Dowdy (1944) referred to the seasonal migration of soil fauna in response to temperature whilst Pierrard, Bonte & Baurant, 1963; Biernaux & Baurant, 1964a and Blower 1970, have specifically monitored the vertical movements of millipedes in either arable or woodland habitats. Barlow (1958) found that temperature rather than rainfall was probably responsible for defining the activity periods of millipedes in a dune environment and that it may also be influential in the initiation of changes in activity during the year. The activity of Oxidus gracilis, a millipede pest of glasshouse-grown cucumber crops (Edwards & Gunn, 1961), was observed by Cloudsley-Thompson (1952) who attributed the appearance of large numbers of individuals on the soil surface at night to an increased activity level caused by a drop in temperature.

There is little evidence in the literature of detailed laboratory work on the behaviour of millipedes to changes in temperature which would seem to be an important factor in governing the amount of damage to crops. In the present study millipedes were subjected to both constant and changing temperatures within observation chambers, whilst humidity was maintained constant, in order to follow both their horizontal and vertical movements. Whether germinated sugar-neet seed can affect their normal responses was also studied.

7.2.2 Materials and Methods

7.2.2.1 Horizontal activity

Five B.superus adults were introduced into a glass observation chamber 6.3 x 0.5 in. (Fig.30). The ends were sealed with rubber bungs, one of which was fitted with a thermistor probe which protruded into the inside of the chamber and which was connected to a Wheatstone bridge circuit to measure temperature. A strip of filter paper (0.05 in. thick) which formed a runway, was moistened to provide a moist surface with a large surface area to give a high level of humidity. The millipedes were left for 4 hours in the tube at laboratory temperature to become acclimatised; after this time condensation appeared on the inside of the glass indicating that the air was saturated with water vapour. The glass chamber was immersed horizontally in a larger glass vessel containing water which was heated by an element and the water circulated by a small propeller (Fig.31). When cooling was required the glass chamber containing the millipedes was raised to near the water surface and placed in a Perspex box 9.9 x 2.0 x 2.0 in. containing water, the temperature of which could be lowered and controlled by introducing either cold tap water or variable quantities of ice. The graduated scale fastened alongside the chamber was used to measure the amount of movement of the millipedes which was timed with a stopwatch. Activity was translated into a comparative figure by ascribing an Activity Index score: the distance moved in cm. by one millipede in a 30 second period was measured at intervals of from 2 to 10 minutes.

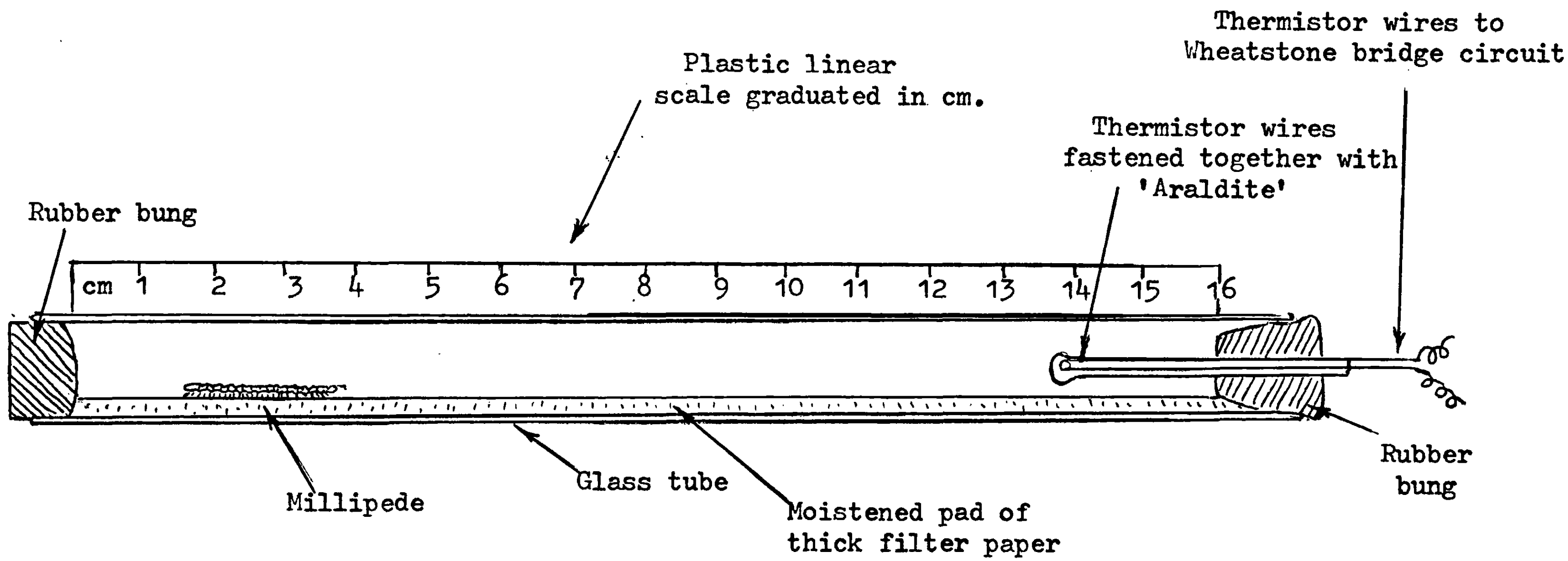


FIG. 30. AN OBSERVATION CHAMBER FOR STUDYING MILLIPEDE ACTIVITY (cross section x 1)

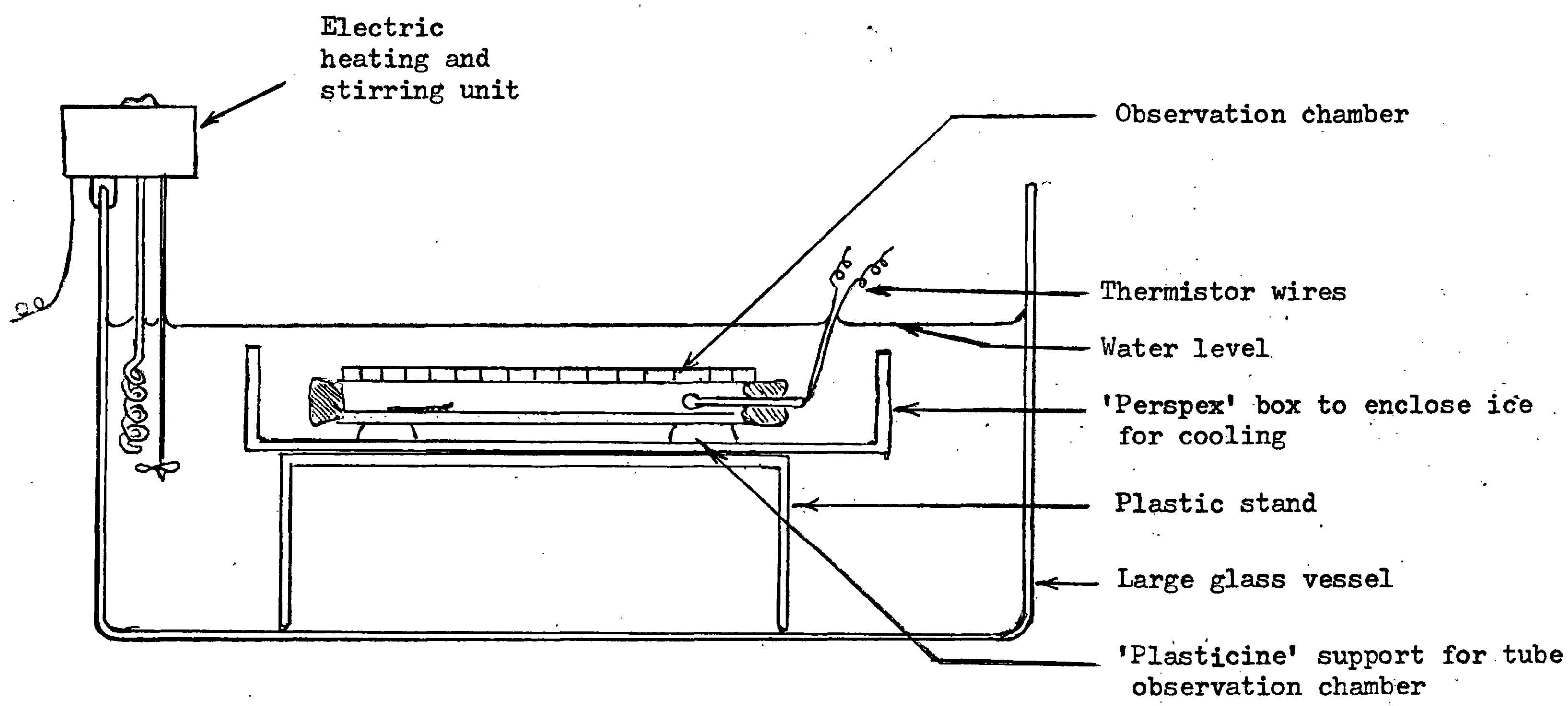


FIG. 31. APPARATUS FOR STUDYING THE EFFECT OF TEMPERATURE ON THE HORIZONTAL ACTIVITY OF MILLIPEDES

(c.s. x 1/3)

A value was obtained for each millipede simultaneously and the total value recorded. The recordings were repeated using 10 fresh B.superus then 5 B.guttulatus.

7.2.2.2 Vertical activity

The same observation chamber was used as in the previous experiment but the filter paper runway was replaced with a 0.4 in. wide strip of compacted moist soil. Ten adult B.guttulatus were introduced into the chamber which was left at laboratory temperature for the air inside to become saturated with water vapour. The apparatus was then arranged as in Fig.32 with the chamber supported vertically within a Perspex box, 7.9 x 2.0 x 2.0 in. fitted with a lid through which the thermistor leads passed. The outside of the chamber was marked with wax pencil into four equal sectors. The water in the glass vessel was heated and stirred by the attached unit whilst the glass observation chamber could be cooled to 2°C by adding cold water or ice blocks to the water in the Perspex box. At intervals of 2 to 10 minutes the Activity Index score for the 10 millipedes was calculated and the number of millipedes in each of the four sectors of the glass chamber was recorded.

7.2.2.3 Vertical activity in the presence of germinated sugar-beet seed.

The glass observation chamber used in the two previous experiments was marked into four equal sectors but the top one was partitioned by a plastic gauze with holes at least 0.1 in. diameter through which millipedes could pass. Ten adult

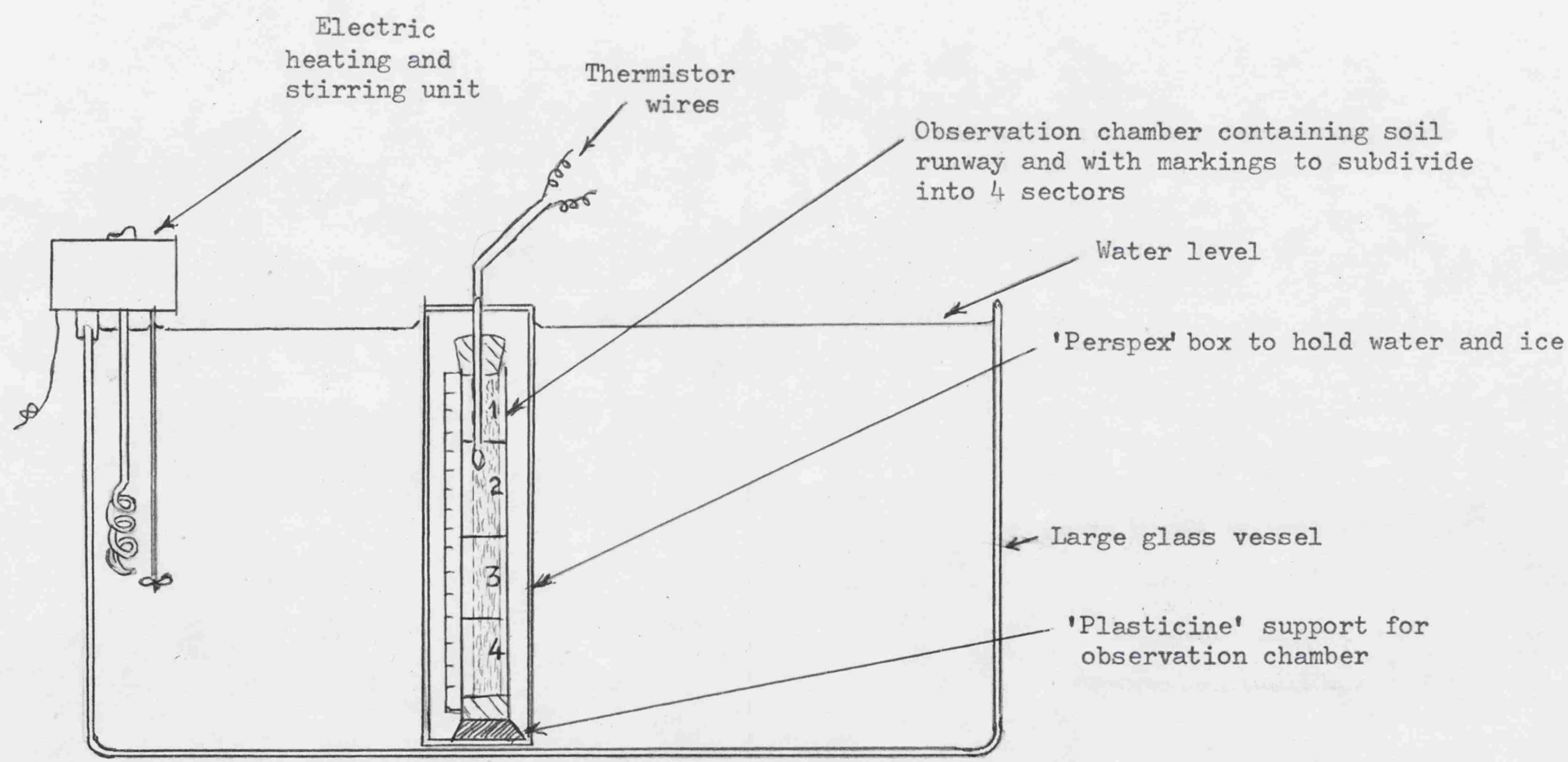


FIG. 32. <u>APPARATUS FOR RECORDING THE EFFECT OF TEMPERATURE ON THE VERTICAL ACTIVITY OF MILLIPEDES</u>

(<u>c.s. x 1/3</u>)

B.guttulatus were introduced and left for 24 hours at a constant temperature. Two, germinated, raw, sugar-beet seed with radicles 0.2 in. long were placed on the plastic gauze at the top of the chamber which was then immersed vertically in the water bath with the seed uppermost. The recordings of activity and position of the millipedes in the tube were started when the temperature in the tube was constant. An Activity Index was calculated as in the other experiments.

7.2.2.4 Millipede aggregation around germinated sugar-beet seed.

A glass-sided wooden box, 12 x 8 x 3 in. was constructed with a front of plate glass and a removable wooden cover which fitted into slots in the front of the box to eliminate light. The box was filled with moist peaty loam soil and ten pre-germinated sugar-beet seed were placed in the soil at a depth of 0.5 in. against the glass front so that all the seed were visible. A thermistor probe was inserted into the soil through an aperture in the side of the box to measure the soil temperature. 150 *B.guttulatus* were introduced into the soil and the lid replaced. To produce temperature variations the box was kept either in a heated laboratory, an unheated room or a refrigerated cabinet. The millipedes around the seed were visible through the glass front and were counted at intervals throughout the day. The wooden cover fitted over the glass to keep out the light was removed for the shortest possible period and the counts performed under low light intensity in order to limit the disturbance since *Blaniulus* is sensitive to light. Up to six counts were performed within the period from 9.00 a.m. until 4.00 p.m., but sometimes it was limited to only one. The observations were continued for one month, after which the seed had become too etiolated for further observation.

7.2.3 Results

7.2.3.1 Horizontal activity

Figs. 33 and 34 show how constant and changing temperatures affect the locomotory activity of the two species. For Blaniulus the intensity of activity is closely related to temperature, the millipedes being more active at 25°C than at 5°C. The effect of constant temperature could not be assessed as this was not maintained for a sufficiently long period. Brachydesmus was less active than Blaniulus and showed little activity at temperatures of less than 5°C; activity declined to zero at constant temperatures and was stimulated by either rising or falling temperature.

7.2.3.2 Vertical activity

Fig.35 shows how temperature changes may affect direction, as well as intensity of movement. Rising temperature caused the millipedes to disperse in an upwards direction as a result of increasing activity and a proportion of the millipedes reached the top sector of the vertical chamber. In response to a lowering of the temperature to 2-3°C there was a net downward movement until all the millipedes came to rest in the bottom of the chamber.

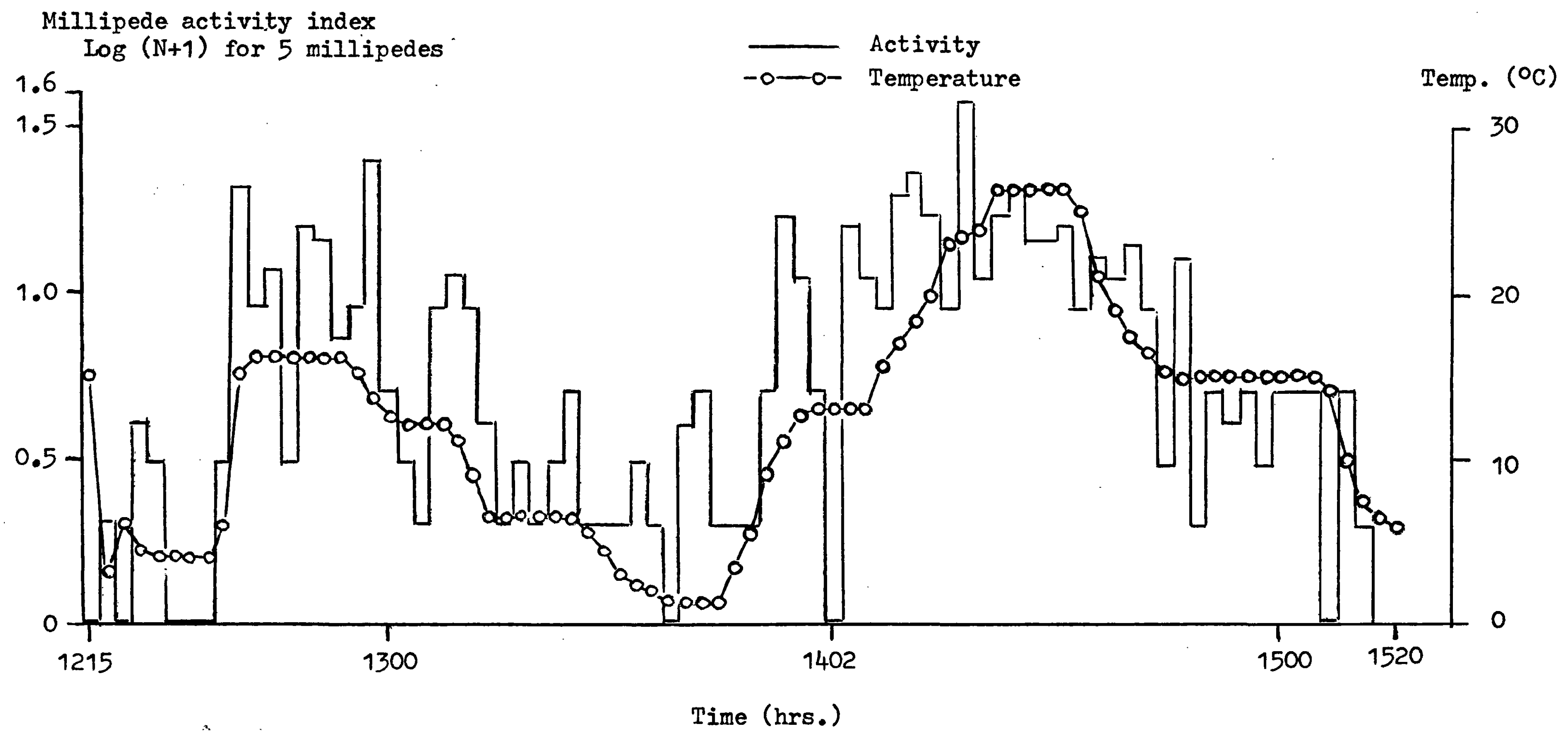


FIG. 33. EFFECT OF TEMPERATURE ON THE HORIZONTAL ACTIVITY OF *BLANIULUS GUTTULATUS*

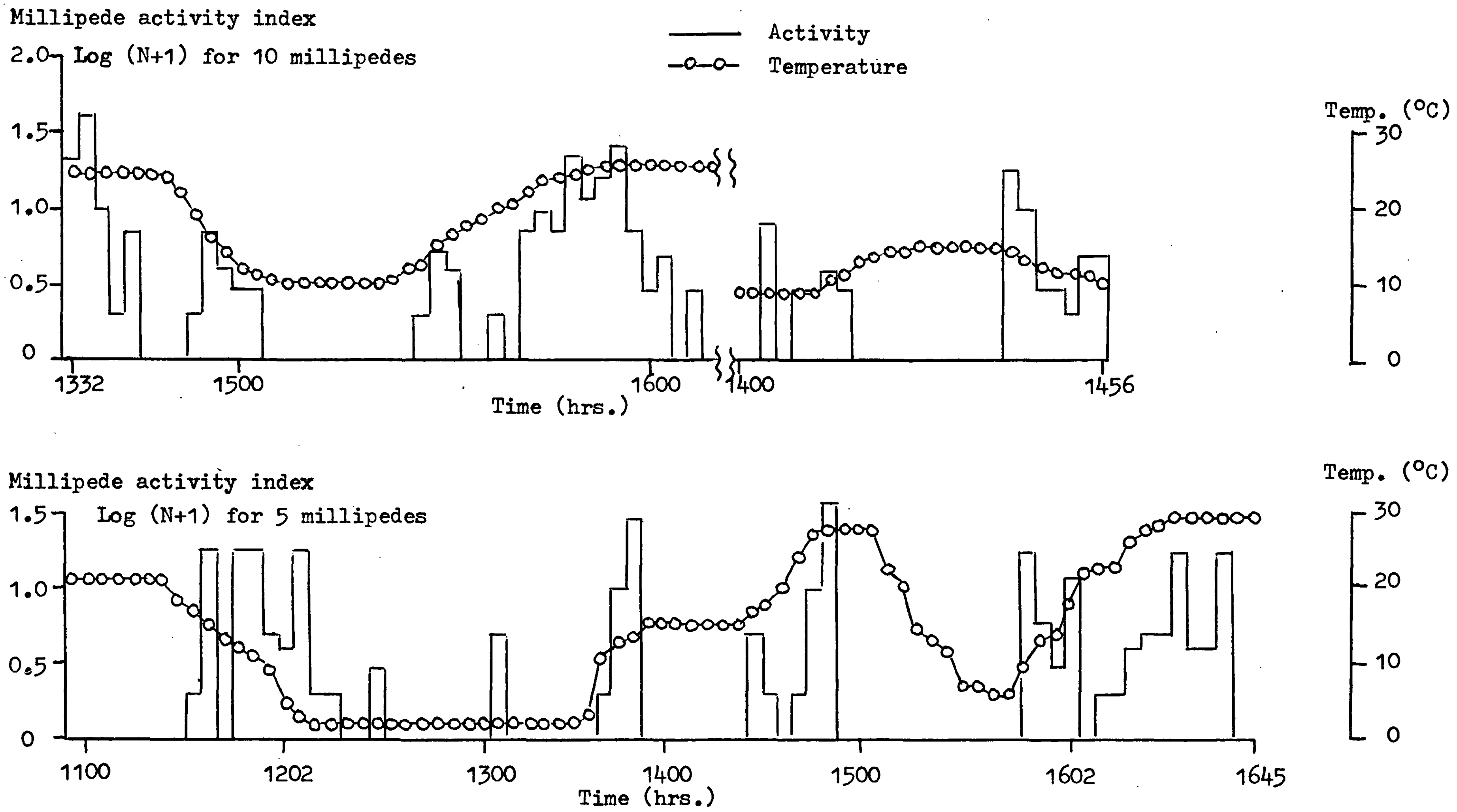


FIG. 34. EFFECT OF TEMPERATURE ON THE HORIZONTAL ACTIVITY OF *BRACHYDESMUS SUPERUS*.

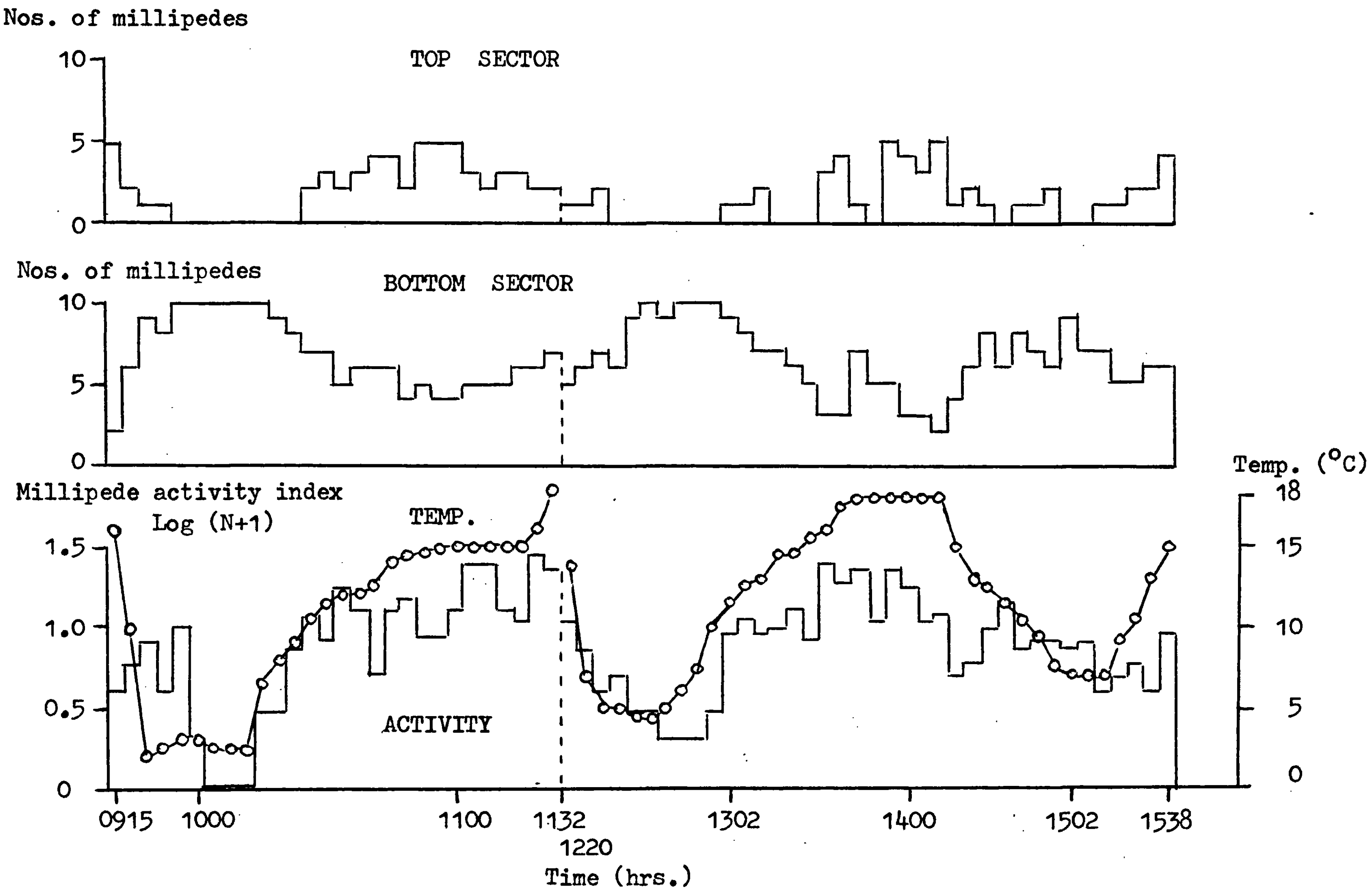


FIG. 35. EFFECT OF TEMPERATURE ON THE VERTICAL ACTIVITY OF BLANIULUS GUTTULATUS

7.2.3.3 Vertical activity in the presence of germinated sugar-beet seed.

In the previous experiment it was seen how falling temperature resulted in a downward movement of millipedes. Fig.36 shows that germinated sugar-beet seed, which are attractive to millipedes, do not prevent this downward movement. In this experiment the chamber was kept at constant temperatures for long periods and the millipedes remained inactive. A resumption of activity was initiated by a temperature change.

7.2.3.4 Millipede aggregation around germinated sugar-beet seed.

Fig. 37 shows the results of a series of 18 one-day observations. On each day the number of millipedes seen around the seedlings varied following a change in soil temperature. As in other experiments the millipedes moved deeper into the soil away from the seedling root zone when the temperature was lowered to 1.5^{o}C. After being maintained at this temperature for approximately 17 hours overnight, the millipedes moved up through the soil and began to aggregate around the seedlings again when the soil temperature was raised to 6^{o}C on one occasion (point A) and 8^{o}C on a similar occasion (point B). After one month the seedlings were no longer attractive but all were damaged by the millipedes.

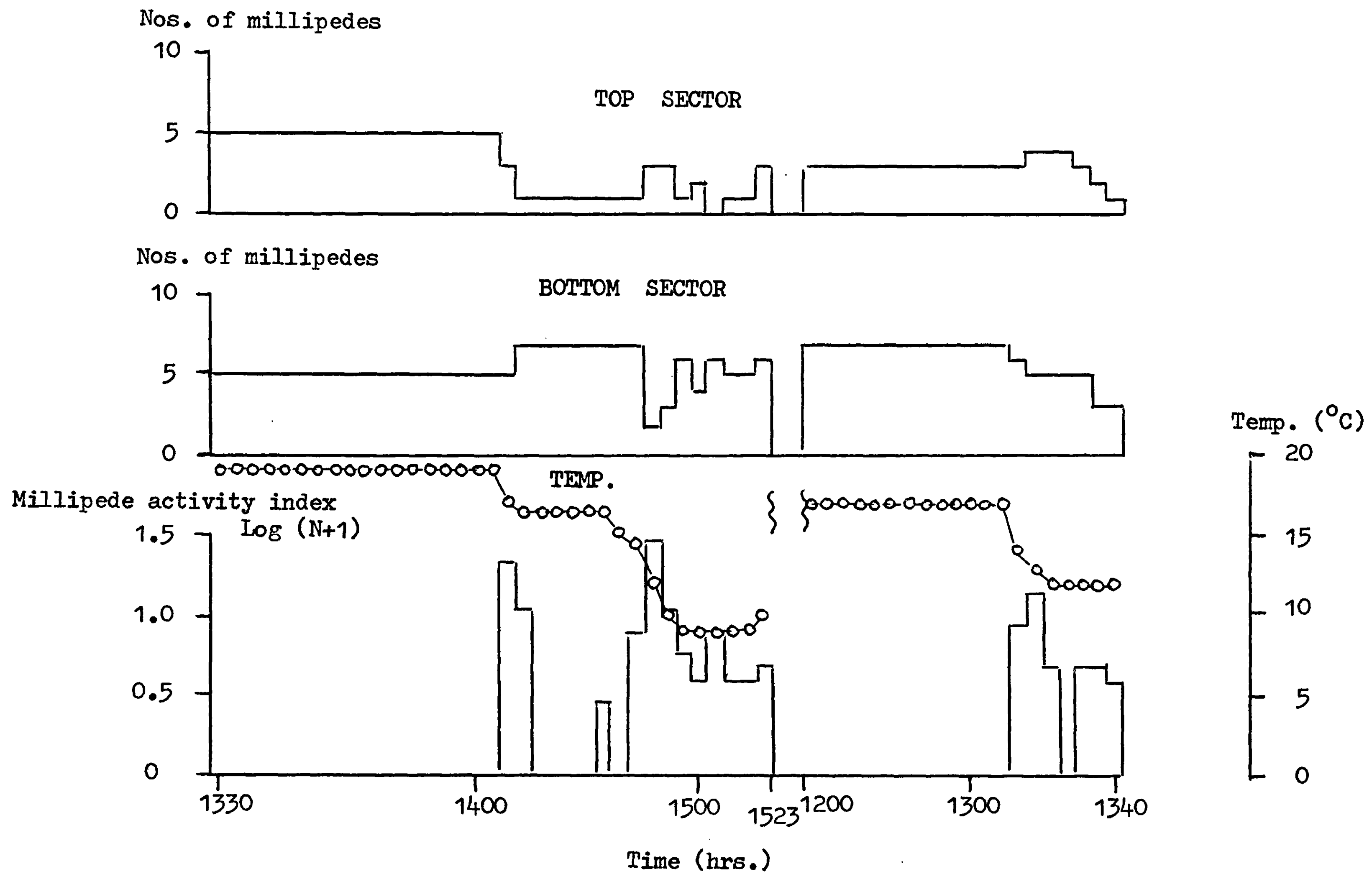


FIG. 36. EFFECT OF TEMPERATURE ON THE VERTICAL ACTIVITY OF *BLANIULUS GUTTULATUS* WHEN GERMINATED SUGAR-BEET SEED ARE CONTAINED IN THE TOP SECTOR OF THE BOX.

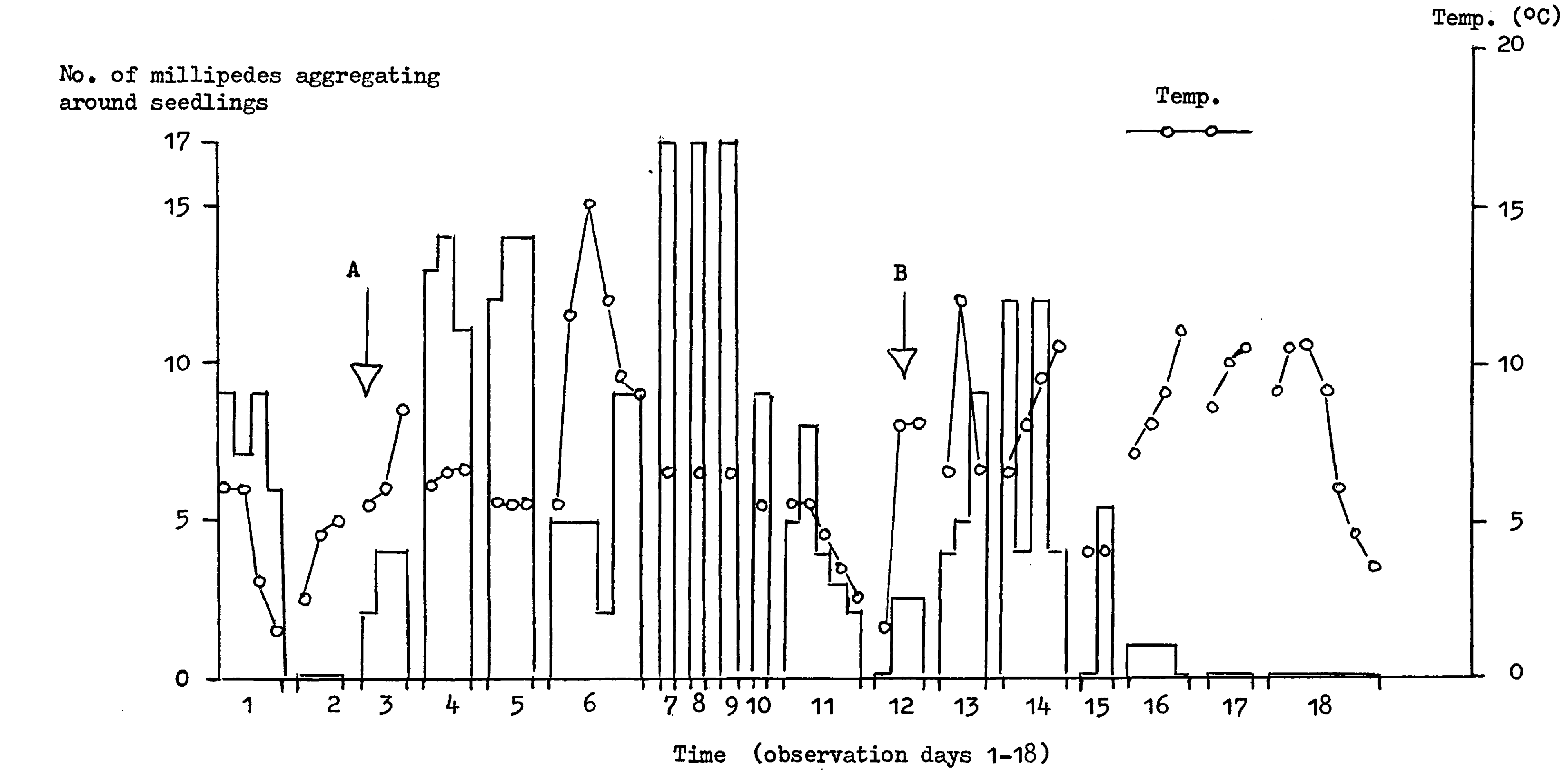


FIG. 37. EFFECT OF TEMPERATURE ON THE AGGREGATION OF *BLANIULUS GUTTULATUS* AROUND SUGAR-BEET SEEDLINGS.

7.2.4 DISCUSSION

The results show that for Blaniulus particularly, temperature may affect both the intensity of locomotory activity and its direction. Locomotory activity increased with temperature and a distinct downward movement was initiated by a temperature fall until, at 2.5°C, all the millipedes had reached the lowest point in the observation chamber, increasing temperature caused a corresponding increase in activity and an upward movement. Bocock & Heath (1967) found that Glomeris marginata (Villers) began to feed and migrate upwards in soil when the daily mean temperature of its surroundings reached 5.5-6.3°C; with a drop in temperature to 6°C in Autumn Glomeris moved downwards and began to eat mineral soil. In the winter, at 2°C and less, downward migration was complete and feeding minimal. In the present studies using the observation chamber, periods of low temperature were not of sufficiently long duration for an accurate description of how activity was affected, but generally there was either no movement or very slight activity below 3°C. Brachydesmus showed little activity below 10°C but periods of rest were broken by both an increase and decrease in temperature.

For millipedes, such as Blaniulus and Brachydesmus which damage sugar-beet seedlings in April and May, it is important to know how spring soil temperatures affect activity and at what temperature they become active and move up into the seed bed to start feeding on the seedlings. These results suggest that upward movement, which is initiated by rising temperature above 0°C, is only a component of a rise in a general level of activity. Blaniulus began to aggregate around sugar-beet seedlings, after a long period at low temperatures, when the soil had reached at least 6°C. From field observations Pierrard, Bonte & Baurant (1963) concluded that a temperature between 4.5 - 5°C was needed to start the upward migration of Blaniulus from deeper in the soil whilst Biernaux & Baurant (1964a) established that the corresponding figure of Archeboreoiulus pallidus was 5.0 - 5.3°C.

Where Blaniulus had been left to aggregate and feed upon the seedling roots a dispersal away from the root zone could be brought about by lowering the temperature sufficiently; the presence of seedlings did not appear to affect their normal directional responses to a temperature fall. Peitsalmi (1974) observed that the direction of vertical movement of the blaniulid Proteroiulus fuscus is dependent upon Relative Humidity. In a uniformly saturated atmosphere all individuals collected in the upper part of his apparatus, whilst in dry air they tended to collect in the lower part. In the present study variations in Relative Humidity, which is temperature controlled, was avoided by enclosing the millipedes in a sealed small-bore glass tube containing sufficient moisture to cause condensation on its interior surface. However, as Relative Humidity was not monitored it was possible that slight variations may have occurred over the large range of temperatures used, but it is unlikely that they would be sufficiently large to elicit a response; Peitsalmi (1974) found that P.fuscus adults did not respond to humidity differences of up to 30% during three-hour experiments.

7.3. <u>FOOD SELECTION BY MILLIPEDES</u>

7.3.1. <u>Introduction</u>

The factors influencing the choice of food material by millipedes may be both chemical and physical and although millipedes are polyphagous, they exhibit food preferences which may determine their distribution (Barlow, 1957). Lyford (1943) found that *Diploiulus londinensis* had a preference for certain species of leaves in litter preferring those with a high calcium content. The softness of the tissue is an important factor determining which organic food is eaten since all millipedes have relatively weak mouth-parts (Brade-Birks, 1930). Cloudsley-Thompson (1950) suggested that millipedes 'may be compelled to attack growing plants particularly during dry periods for the sake of moisture' and that 'sugars in the plant sap restrain them from returning to their normal diet of humus and decomposing vegetation'. Although acknowledging that millipedes will attack growing crops, Cloudsley-Thompson relegates millipedes to the position of secondary feeders since he states that other pests such as wireworms must make the initial entry. Whilst millipedes often accompany other pests, observations in the laboratory show that they can initiate the attack on sugar-beet seedlings. Rotting substances, on which millipedes are frequently found, such as potato tubers (Carpenter, 1910; Rolfe, 1937; Vachon 1942) have a high sugar content brought about by the release of simple sugars from the breakdown of polysaccarides by fungi and bacteria. Litter leaves, on which millipedes feed, also contain similar sugars (Sakwa, 1974).

It is possible that millipedes prefer to feed on sugar-beet seedlings because of their sugar content, but the main purpose of the present study was to determine which sugars, from a small range, are most attractive and to combine it in a water-agar to test the hypothesis that 'dry' conditions may cause millipedes to attack plants for the sake of moisture. The sugar-agar was tested as a seedling substitute since it would be more amenable to gravimetric measurements and present no physical barrier to millipede feeding. Moreover, agar jelly may contain about 90% water which approximates to that contained by succulent living plant tissue (Curtis & Clark, 1950).

7.3.2. A preliminary experiment

Whether a dilute sugar solution dissolved in water agar was attractive to Blaniulus guttulatus was tested and the suitability of agar as a carrier medium was assessed.

A 2% water-agar (control) and a 2% water-agar containing a 2% solution of glucose was each poured into two separate Petri dishes to a depth of 0.1 in. Eight agar discs were cut from each using a 0.5 in. diameter metal auger and the discs from the two agars arranged alternately around the circumference of the two Petri dishes lined with moist filter paper. To maintain a high humidity and provide a suitable environment for the millipedes, the Petri dishes were placed on top of each other upon a layer of moist vermiculite in a box with a snap-fitting lid. After the addition of 10 Blaniulus guttulatus to each Petri dish the apparatus was left in a dark cupboard at laboratory temperatures. At 24 hr. intervals the number of bites on the top surface of each disc was counted and fresh discs introduced. After three days the agar discs containing the 2% glucose had an average of 63 bites per disc and the control, without the sugar, 3 bites per disc. The number of bites on each glucose-agar disc varied from 4 to 148 implying that Blaniulus was feeding in groups on one disc for long periods. This exercise established that Blaniulus will readily feed on agar to which glucose had been added but the method of assessment was difficult and many discs were eaten round the side leaving large holes where the number of bites could not be assessed; some shrinkage was also evident.

7.3.3 Materials and Methods

7.3.3.1. Sugar preference of Blaniulus guttulatus.

Petri dishes containing 5% water-agar plus 5% solutions of each of fructose, sucrose and glucose plus a control, without a sugar, were prepared. Whilst the agar was still hot and molten, 0.1 in. deep plastic rings, 0.5 in. internal diameter and prepared from plastic piping, were placed in the agar which was approximately 0.2 in. deep. The object was to prepare agar discs of uniform size and with sides enclosed by the plastic ring in order to minimise water evaporation and facilitate handling. When the agar had solidified the plastic rings enclosing the agar discs were removed from the parent medium and surplus agar was trimmed away leaving discs of exactly 0.1 in. thickness; the weight of each disc was recorded. Eight glass Petri dishes containing a 0.2 in. layer of moistened Plaster of Paris were prepared, each with 4 equally spaced circular holes in which the plastic rings could be fitted. Into each hole was first poured a 0.1 in. layer of plain agar to provide a base; this was allowed to solidify; then four plastic rings, each containing different agar discs, were placed in each Petri dish in these holes. The agar within the plastic rings was now level with the surface of the plaster block. A small quantity of soil was placed on the surface of the plaster in each Petri dish. Forty Blaniulus guttulatus were introduced into each Petri dish and the lids secured with adhesive tape. The 8 Petri dishes were kept in a dark cupboard at laboratory temperatures (15°-20°C). After 1, 4 and 5 days the plastic rings, each enclosing an agar disc, were removed with forceps and weighed. After 5 days fresh discs were prepared and the experiment was continued for a further 2 days after which the final disc weight was recorded. After every weighing the millipedes were removed and placed together in a single container; they were reallocated to each Petri dish at random before continuing the experiment.

7.3.3.2. Influence of soil moisture on agar consumption by Brachydesmus superus.

Petri dishes containing the following agars, approximately 0.1 in. deep, were prepared :-

5% water-agar (control)

5% water-agar plus 5% glucose

5% water-agar plus 5% sucrose

A 0.4 in. diameter cork borer cut out 3 discs from each agar; these were weighed and each placed on separate 0.5 in. square microscope cover slips on the soil surface of each of three Petri dishes. All dishes contained a 0.2 in. layer of a mixture of fine sandy loam and peaty loam, offered either 'dry' (8 - 11% m.c.) or 'moist' (24 - 28% m.c.). Thirty adult B.superus were added to each Petri dish which were placed within a polythene box lined with thick moist filter paper to minimise moisture loss. The weight of the agar discs was recorded after 1, 4 and 8 days, after which they were replaced by fresh discs and the experiment continued for a further 3 days. The millipedes were mixed and reallocated to each Petri dish at random as in previous experiments after each disc weighing.

7.3.4. Results

7.3.4.1. Sugar preference of Blaniulus guttulatus.

Fig. 38 shows the relative cumulative consumptions of the agars containing different sugars compared with the control for a seven day period. At every weighing least agar was consumed from the control and most from that containing 5% sucrose; fructose was the least palatable sugar. After five days some slight discoloration of the agar was apparent, probably due to colonization by bacteria since bacteriocide was omitted following the autoclaving of the agars.

7.3.4.2. Influence of soil moisture on agar consumption by Brachydesmus superus.

Fig.39 shows the relative consumptions of both control, glucose and sucrose-agars when offered to B.superus on soils of contrasting moisture status. The weight of agar consumed is a total for each of the three Petri dishes per treatment, each dish containing three agar discs and 30 B.superus. As in the previous experiment, agar containing sucrose was the one eaten most. Generally more agar was consumed in Petri dishes containing the moist soil. A 't' test was used to compare the mean agar consumptions per Petri dish on either 'moist' or 'dry' soil; there was no significant difference ($t = 0.5$ with 16 d.f.).

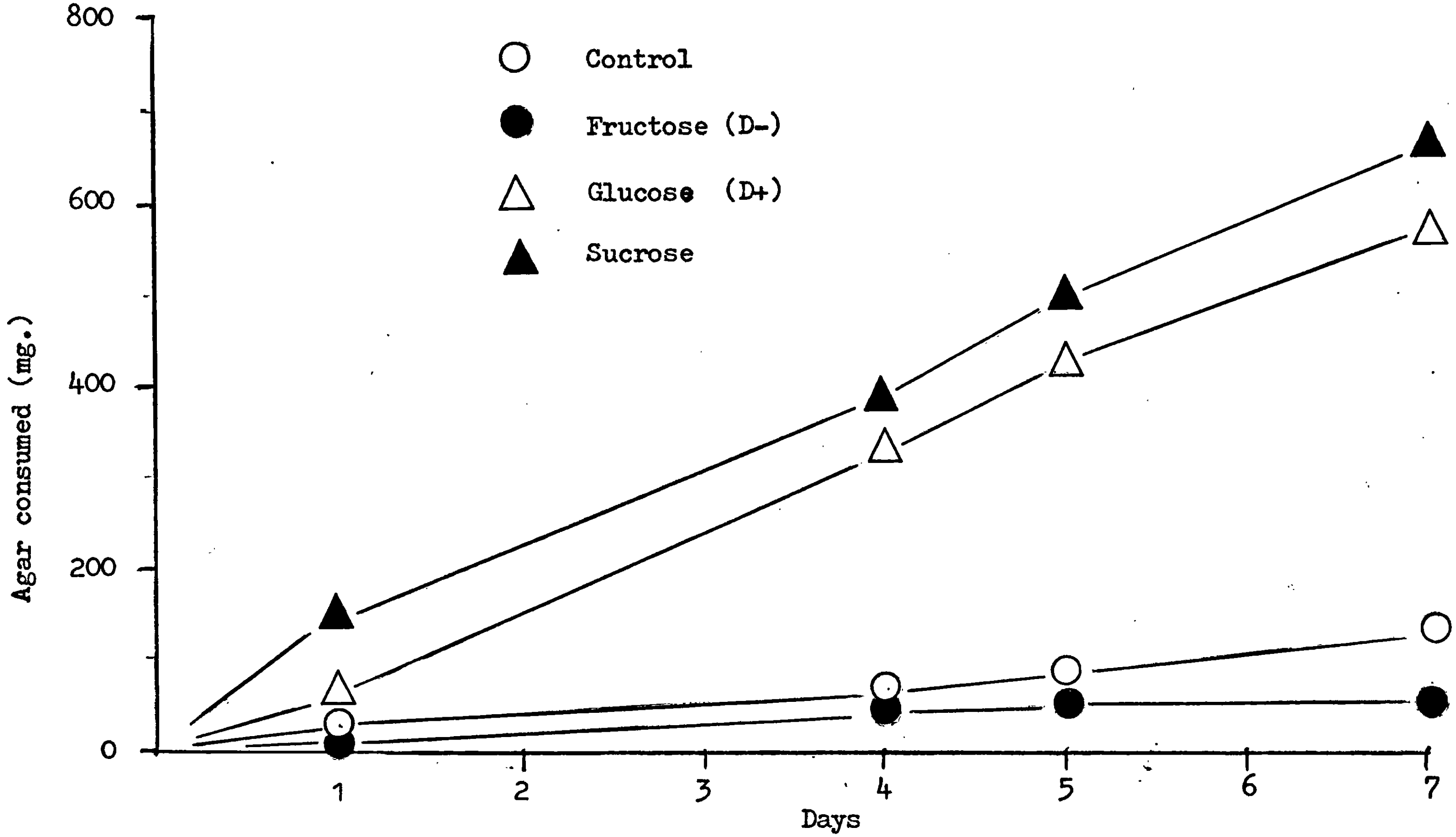


FIG. 38. CUMULATIVE CONSUMPTION OF AGAR CONTAINING DIFFERENT SUGARS BY *BLANIULUS GUTTULATUS*

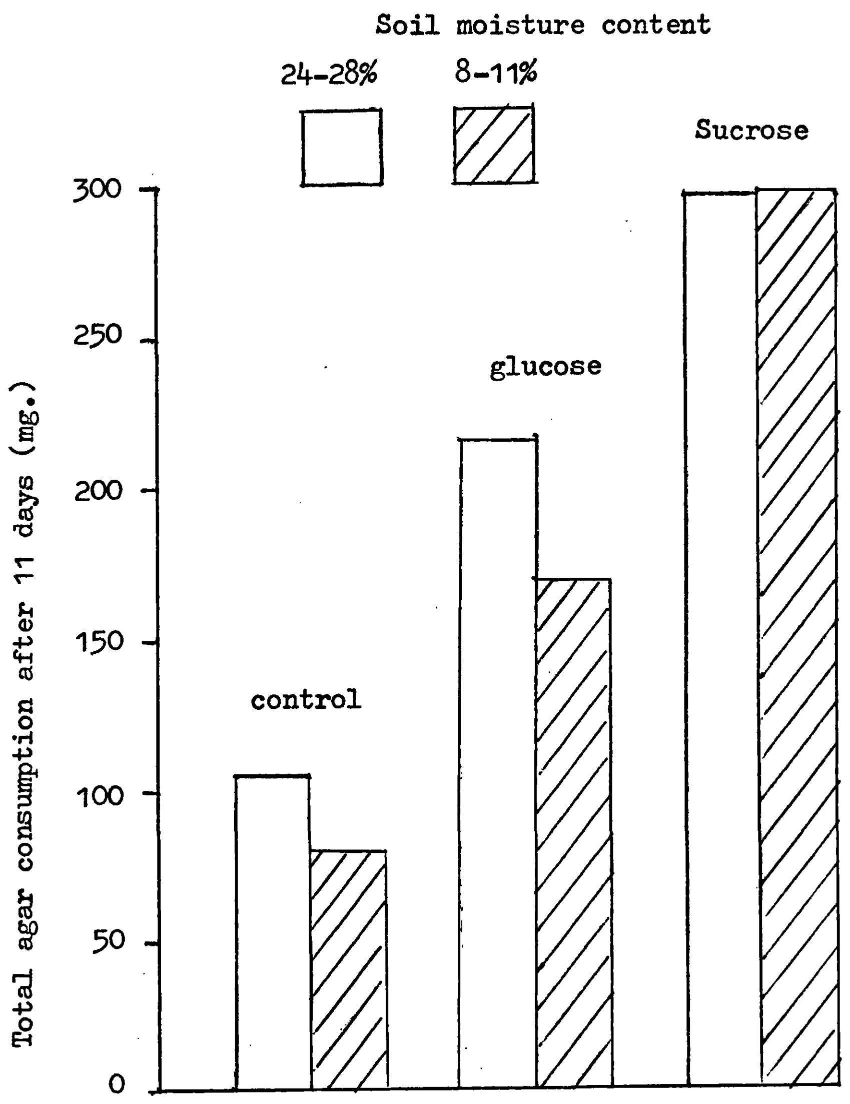


FIG. 39. <u>EFFECT OF SOIL MOISTURE CONTENT ON CONSUMPTION OF AGAR BY BRACHYDESMUS SUPERUS</u>

7.3.5. Discussion

In the experiments both B.guttulatus and B.superus showed a general preference for agar containing sucrose rather than glucose or the control agar not containing a sugar. Cloudsley-Thompson (1951) found that B.guttulatus and Paradesmus gracilus were attracted to filter paper soaked in 2% solutions of glucose and sucrose and to air-dried humus soaked in solutions of 1% and 0.05% glucose and 0.05% sucrose; they were also repelled by 2% urea. Urea is a common agricultural fertiliser and when part of the hydrogen is replaced with other elements, substituted ureas are formed, which are herbicides. Although not tested in the present study, it would be interesting to investigate the effect of ureas on the distributions of B.guttulatus in the field.

The consumption of agar on soils of contrasting moisture content did not differ significantly. The hypothesis that 'millipedes will be compelled to attack growing crops for the sake of moisture' (Cloudsley-Thompson,1950) was formulated from the high incidence of reports that, where a millipede attack on a crop occurred, soil conditions were usually dry. In dry conditions seedlings will be under a moisture stress, particularly if the roots have not penetrated very deep. When a seedling is deprived of water growth is restricted and because it can neither mature enough to harden its root tissue against millipede attack, nor compensate by growing replacement water or nutrient-absorbing tissue, the seedlings would therefore be expected to succumb more easily to a given intensity of millipede attack under 'dry' soil conditions.

In the field, wilting seedlings that are being attacked by millipedes are common when the soil is dry and the seedlings small. Adjacent seedlings, free of millipede damage, are able to grow normally and extend their root systems into deeper more moist soil. The attacked seedlings usually die in dry conditions but may recover only if the soil is re-moistened through the growth of lateral roots from the damaged areas. Therefore, it is probable that loss of seedlings through millipede damage is aggravated by dry soil conditions through an effect on growth rather than intensity of millipede numbers. It has not been possible to simulate these conditions in the laboratory using seedlings.

8. SOME FACTORS WHICH MAY EFFECT THE DAMAGE TO SUGAR-BEET SEEDLINGS BY SOIL-INHABITING PESTS.

8.1 FEEDING STUDIES ON MILLIPEDES AND COLLEMBOLA

8.1.1 Attractiveness of raw and pelleted seed

8.1.1.1. Introduction

In commercial growing of sugar beet the use of pelleted seed is now almost universal whereas in 1965 it represented only 3% of the total acreage in Britain. The transition to pelleted seed has paralleled the increase in precision drilling, herbicide usage and use of genetic monogerm seed (Hull & Jaggard, 1971). In feeding experiments with pests, therefore, it is useful to test whether pelleted seed, not treated with insecticide, has lessened or increased the germinated seeds' susceptibility to pest attack, particularly before the cotyledons have emerged above the soil surface. In the present studies both types of seed were offered to blaniulid millipedes. Later, in the laboratory, it was tested whether a subterranean species of Collembola (*Onychiurus* spp.) could damage sugar-beet seedling roots and the amount of damage was compared to that caused by millipedes under similar conditions. An experiment in the field, where *Onychiurus* were numerous, tested the numbers around seedlings grown from raw and pelleted seed, root damage and seedling establishment.

8.1.1.2. Materials and Methods.

8.1.1.2.1. Observations on millipedes in the laboratory.

Rubbed and graded sugar-beet seed, both in raw and pelleted form, (cultivar 'Amono') was left to germinate on moist filter-paper. The seeds were removed and used in the experiment when the radicles had begun to emerge through the opening in the fruit. Ten seeds of each type were placed in rows on the surface of heat-sterilized moistened, loose soil in a polythene box (7.0 x 4.9 x 1.8 in.) with an airtight lid. One hundred and fifty adult Boreoiulus tenuis were chosen at random from a culture kept at 5-10°C and placed in the box with the seeds; the same millipedes were used throughout. The box was left in an unheated room where the temperature varied from 9°C to 12°C over the duration of the experiment.

The number of millipedes around each seed was counted and recorded at three separate 24 hr. intervals; after three days the seeds were removed (the root was now approximately 0.5 in. long) and the amount of damage to each seed assessed by a scoring method with a scale of 0 - 5 (0 = seedling free from damage; 5 = damage sufficient to cause death of seedling - extensive feeding lesions on cotyledons, hypocotyl and/or radicle). For the second sowing pre-germinated seed of both types were again placed on the surface of the soil and observations carried out as described. The experiment was continued for two months; this was the maximum period that the millipedes would cause damage to the seedlings representing a total of six consecutive sowings. The differences in mean aggregation and damage between the two seed types was tested by a 't' test.

8.1.1.2.2. Observations on millipedes and Collembola in the laboratory.

Both raw and pelleted sugar beet seed (cultivar 'Monotri') were pregerminated in the laboratory until the radicles were visible, as in the previous experiment. Two polythene boxes (7.0 x 4.9 x 1.8 in.) were each half-filled with moistened, heat-sterilized, silty loam soil. One hundred and eighty adult Blaniulus guttulatus were introduced into one box and 180 Onychiurus armatus into the other from culture stocks maintained at 7°C. Two rows of 5 raw and two rows of 5 pelleted seed were placed in small depressions in the soil in each box, the lids replaced and the boxes stored at 7°C. For each sowing the number of millipedes or Collembola around the seed was counted every 24 hrs. After 6 consecutive 24 hr. periods the seeds were carefully removed, washed on a sieve to remove soil and each scored for damage on a 0-5 scale, as in the previous experiment. After a total of 8 consecutive sowings, over a period of three months, the boxes were emptied and the number of millipedes and Collembola remaining were counted to determine percentage mortality. Significance of the mean millipede numbers and mean damage per seed was tested by Analysis of Variance treating each sowing as a replicate.

8.1.1.2.3. A field experiment.

A site at Stalham (Norfolk) in 1974 showed a population of between 7 and 8 million O.armatus per acre in March and April, sampling to a depth of 7.8 in. An experiment was planned to test both raw and pelleted seed, not treated with an insecticide and two insecticide treatments incorporated into the pellet. The insecticide effects are reported in the section on the 'Control of soil-inhabiting pests of sugar-beet seedlings by insecticides' and will not be described here. Single row plots, 63 ft. long, were laid out in six randomised blocks. Each block had the two treatments (raw and pelleted seed) randomised four times. The seed, (cultivar 'Monotri') was drilled at 3 in. spacing for the pelleted seed and 3.3 in. for the raw seed on 10 April (the slight difference in seed spacing was unavoidable due to the belt and drive combinations in the seed drill units). The centre row of each 5-row 'plot' was a discard row drilled at 3 in. spacing with pelleted seed. On 29th April, before the cotyledons had emerged above the soil surface, a one yard length of row to a depth of 3 in. was removed at random from each single row plot on the three blocks of the experiment where most _Onychiurus_ were found; this approximated to 5 lbs., or 2 litres, of soil per plot. From each plot it was placed in a separate labelled polythene bag and brought back to the laboratory where the seedlings were removed, washed on a sieve and scored for root damage on a 0-5 scale. The _Onychiurus_ were extracted by crumbling the soil over a Ladell vessel full of cold water; The _Onychiurus_ were picked off the water surface with the tip of a fine brush and numbers of both living and dead individuals per yard length of row recorded. The data was analysed by Analysis of Variance.

8.1.1.3. Results

8.1.1.3.1. Observations on millipedes in the laboratory.

Fig.40 shows the number of millipedes observed around each seed type (10 seeds) as a mean of the three 24 hr. observations and the seedling damage at each of the six consecutive sowings. The histograms show that both aggregation and damage were greatest at the initial sowings and declined markedly throughout the experiment until, after 6 sowings, only few millipedes were seen around the seed and damage was very slight, even though the millipedes remained active throughout. More millipedes were also recorded around the pelleted than around the raw seed at every sowing. From all sowings, 74% more millipedes were found around pelleted seed, the difference being significant at $P < 2\%$ ($t = 2.57$). Differences in the amount of damage to the seedlings was very slight and insignificant.

8.1.1.3.2. Observations on millipedes and Collembola in the laboratory.

Fig. 41 shows the comparative aggregation of the pests and damage to raw and pelleted seed by Blaniulus and Onychiurus and represents a mean of six sowings. More Blaniulus were found around pelleted than raw seed but the difference was not significant. Generally more Blaniulus were recorded around seed compared to Onychiurus but the significantly higher number around pelleted seed is not reflected in the pattern

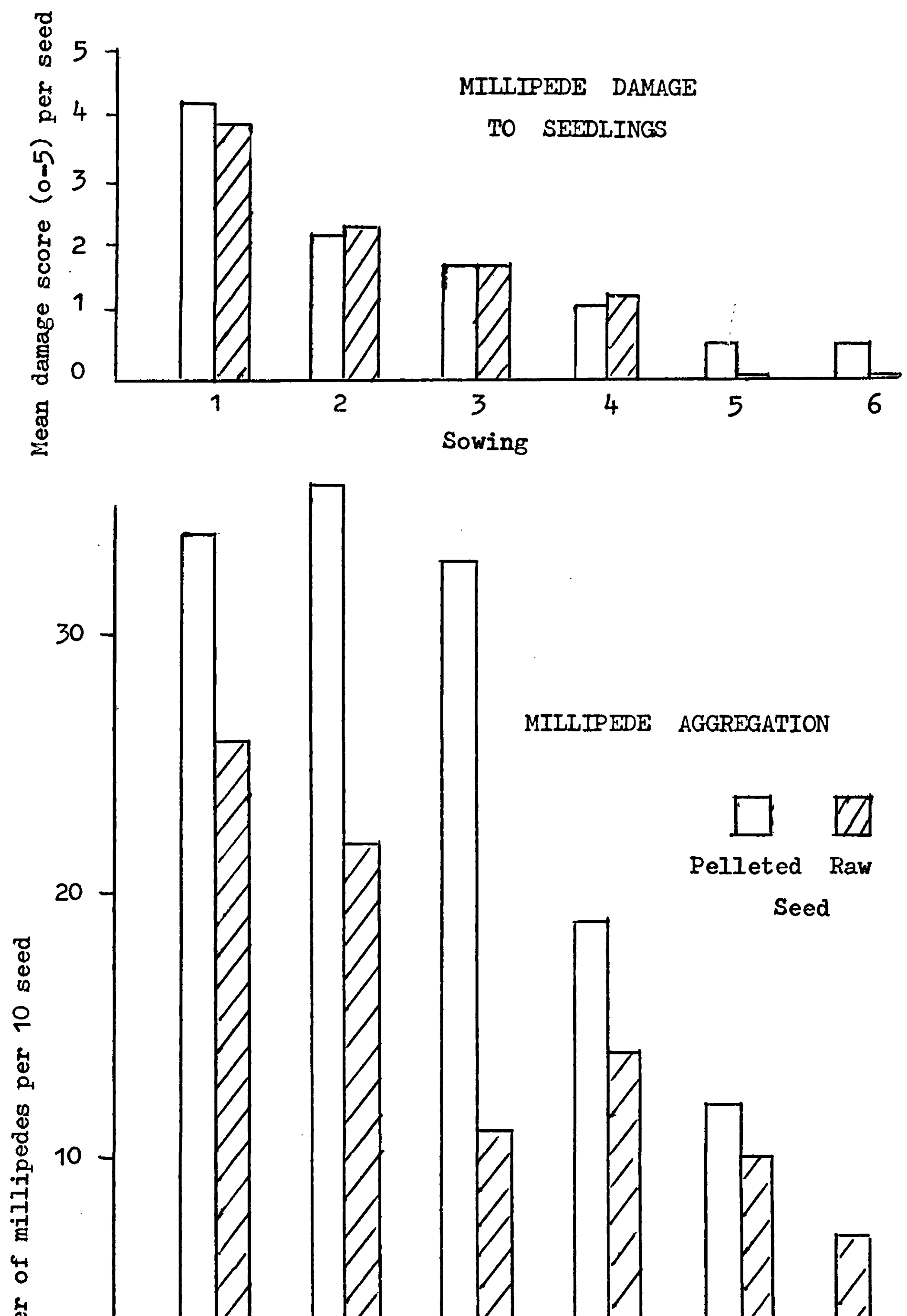


FIG. 40. <u>EFFECT OF RAW AND PELLETED SEED ON MILLIPEDE AGGREGATION AND DAMAGE TO SEEDLINGS</u>.

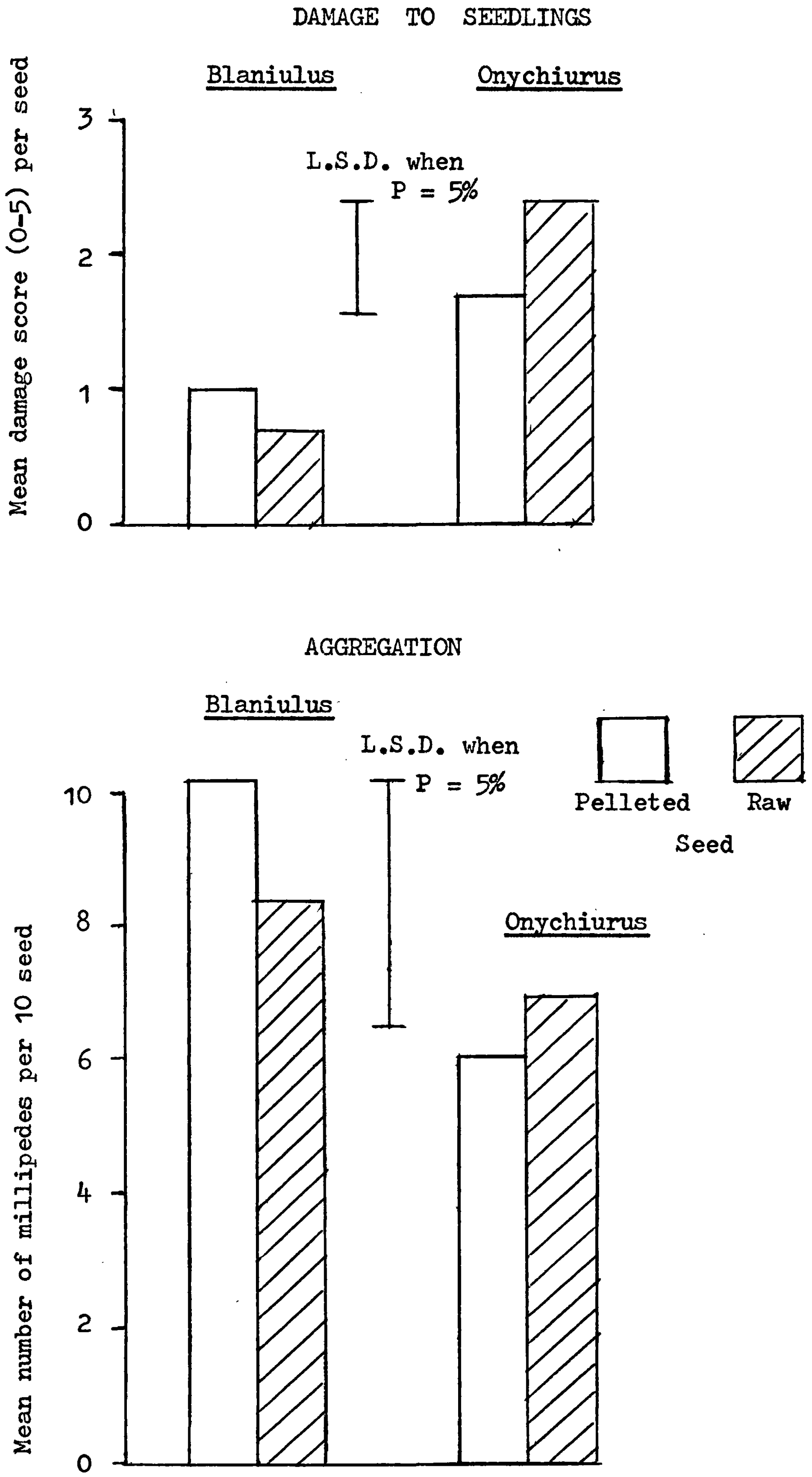


FIG. 41. AGGREGATION AND DAMAGE TO RAW AND PELLETED SEED BY BLANIULUS MILLIPEDES AND ONYCHIURUS COLLEMBOLA IN THE LABORATORY.

of damage to the seedlings; *Blaniulus* was the less damaging of the two pests. *Blaniulus* caused significantly less damage to raw seed than *Onychiurus*. Considering the damage to both types of seed combined, *Onychiurus* was the most damaging; the differences were highly significant, $P < 0.1\%$, (Appendix table 3). At the last sowing *Blaniulus* did not damage any seedlings but *Onychiurus* damaged the roots of many severely. Plate 23 shows some typical damage caused by *Onychiurus* and plate 24 the effect on seedling growth compared to some undamaged seedlings. Of the total number of the pests maxima of only 24% and 15% of the *Blaniulus* and *Onychiurus* respectively were observed feeding at any one time; mortality was 28% and 9% respectively at the end of the experiment after three months.

8.1.1.3.3. A field experiment

The number of seedlings recovered in the yard length of row samples varied for the two seed types, averaging 6.5 for raw seed and 9.1 for pelleted seed; many had not yet germinated when the samples were taken. Table 33 shows the comparative numbers of *Onychiurus* recovered, mean root damage score per seedling and percentage seedling establishment for each seed type. Numbers of the pest around the two types of seed did not differ significantly and variation was large. The seedlings grown from raw seed suffered most damage but this just failed to be significant. Seedling establishment was significantly better ($P < 0.1\%$) for pelleted seed.

PLATE 23. SOME SUGAR-BEET SEEDLINGS GROWN IN THE LABORATORY DAMAGED BY *ONYCHIURUS ARMATUS* x 10.

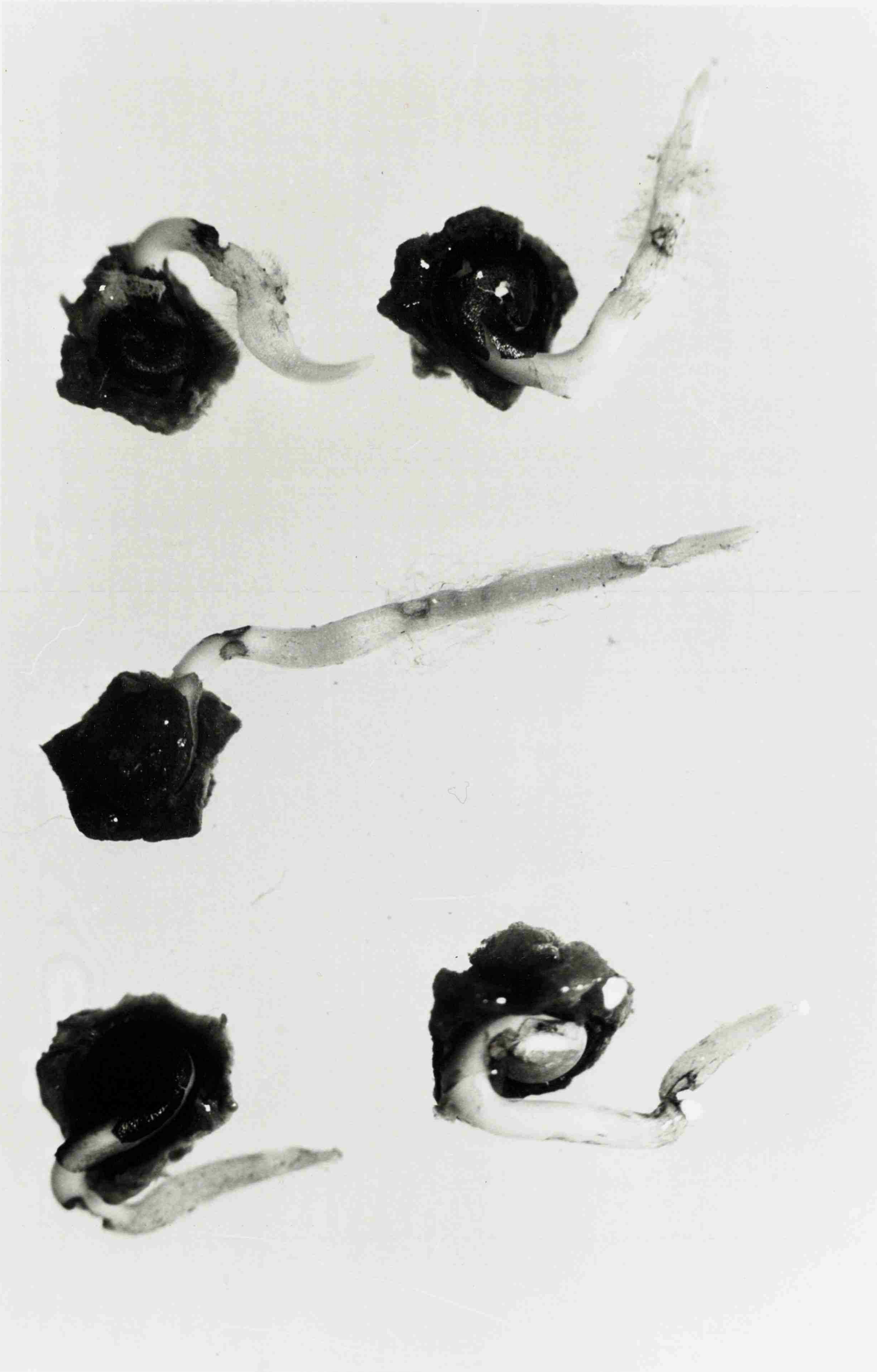


PLATE 24. EFFECT OF DAMAGE BY *ONYCHIURUS ARMATUS* ON THE GROWTH OF 20 SUGAR-BEET SEEDLINGS (TOP TWO ROWS) COMPARED WITH 20 UNDAMAGED SEEDLINGS (BOTTOM TWO ROWS) GROWN IN THE LABORATORY x 2.

TABLE 33. EFFECTS OF USING BOTH RAW AND PELLETED SEED IN A FIELD EXPERIMENT

Treatment	*Onychiurus*/yard of row		Mean root damage score/seedling	Seedling establishment
	Living	Dead	(0 - 5)	(%)
Raw seed	28	12	2.79	37.6
Pelleted seed	41	21.4	2.46	60.5
L.S.D.				
P = 5%*	17.9	12.2	0.5	4.3
P = 1%**	-	-	-	5.8
P = 0.1%***	-	-	-	7.7
P	n.s.	n.s.	n.s.	***

8.1.1.4. DISCUSSION

The results of the laboratory experiments indicate that millipede feeding decreases with time as successive sowings are offered. If this occurs in the field the practical significance is considerable since, even if the seedlings mature only slowly, the amount of fresh damage that the millipedes inflict will become less; also, a redrilling following the ploughing-up of a severely attacked crop will not necessarily become damaged to the same extent as the preceding crop. In the first experiment described the millipedes were more attracted to pelleted rather than raw seed, but other studies have shown that the pelleting material *per se* is not the source of attraction; it was offered in a choice-experiment comparing the different components of germinated seed with pellet material removed, by soaking, from dry seed. Observations in all feeding experiments indicate that aggregation and feeding does not occur until the seeds germinate and the radicle is accessible. In the experiments described the pelleting material may well have absorbed some nutrient leachates from the seed coat, and in this modified way, have served as a focus for the millipedes. In future reference to germinated pelleted seed, the pellet material might be considered as a source of adsorbed leachates from the fruit and as offering an increase in surface area as compared to the raw seed. That the millipedes were probably just as much attracted to this modified pelleting material as to the seedling it enclosed, at one point, is implied in the results since though the seedlings from pelleted seed attracted more millipedes the damage was of equal intensity for both seed types.

In the second experiment, although more millipedes were attracted to the pelleted than the raw seed, the difference in numbers was less pronounced than in the previous experiment and non-significant. Although fewer Collembola were seen around the seed, they caused more damage, but this may have been partly due to their ability to avoid detection by moving into the soil crevices; their small size made observations more difficult than with millipedes. On some occasions many millipedes were observed in contact with the seed but inspection of the seedlings did not reveal any evidence of damage.

The results provide evidence that Onychiurus can be regarded as a serious pest of sugar-beet seedlings and the concept of them as purely secondary feeders is no longer valid. They have caused damage to seedlings grown in the laboratory similar to that detected in the field, leaving distinct feeding lesions, that on close inspection were not likely to be areas of tissue with bacterial or fungal disease. Both Edwards (1962) and Heijbroek (1972) have reported similar damage by Onychiurus spp. to crops. It is perhaps significant that, when present in equal numbers, they can cause as much, or more, damage to sugar-beet seedlings as millipedes, which are established pests. Many cases of damage, or a favourable response to a soil insecticide on crop growth where the common soil-inhabiting pests are absent, may be due to Onychiurus; Morris (1927) found that O.armatus were sometimes the most abundant insect species in the soil.

In the field experiment there were 29% fewer seedlings recovered grown from raw seed than pelleted seed. This cannot entirely be accounted for by the 14% difference in the expected number of seedlings per yard of row attributable to the small difference in respective seed spacings; in laboratory experiments there was no observable difference in their rate of germination or development. A possible explanation of the result could be that many embryos from raw seed were completely eaten before they were sampled and were not recovered from the soil because they failed to develop into recognisable seedlings that could be picked out in the laboratory. The greater susceptibility of the seedlings from raw seed to damage by *Onychiurus* was consistent with laboratory observations.

8.2. EFFECT OF TIME OF SOWING

8.2.1. Introduction

The extent to which sugar-beet seedlings are damaged by millipedes is very variable from year to year (Dunning, 1975) and dependent mainly upon two factors: the seedlings' resistance to attack, which is governed by its rate of growth and the size and activity of the millipede infestation in the seedbed; both these factors are controlled mainly by weather conditions, primarily temperature and rainfall. In spring top-soil temperatures are slowly increasing and usually the crop is sown in March to April, though the earliest sowings in March, when the soil is cold, may take up to a month to emerge (Hull & Jaggard, 1971). The period between germination and emergence of the cotyledons above the soil surface is when seedlings are at greatest risk from soil-inhabiting pests; the shorter this period the greater the chance of escaping pest damage since the seedlings will be growing rapidly and will be best equipped to replace damaged root tissue. By altering the sowing date the rate of germination and emergence of sugar beet can be affected since the soil will be either warmer or colder. In 1972 and 1973, experiments tested the effect of different times of sowing of a sugar-beet crop in fields having a millipede infestation.

8.2.2. Materials and Methods

Both at Shouldham (Norfolk) in 1972 and at Marham (Norfolk) in 1973 blaniulid millipedes, Boreoiulus tenuis, were found in the seedbed in April, averaging 11.5 and 14 million per acre respectively in the top 3.9 in. At both sites the soil was a calcareous very fine sandy loam(with some flints)and millipedes had been recorded causing damage to previous sugar-beet crops. At Shouldham a 4 x 4 randomised block experiment was laid out with plots 8.3 ft. wide (5 rows) and 50 ft. long. Pelleted sugar-beet seed (cultivar 'Amono') was sown at 1.5 in. spacing and 1.5 in. deep at four different times with a three week interval: 20 April, 12 May, 1 June, 23 June. The first was sown with a 'Stanhay' tractor mounted seed drill, later sowings using a single 'Stanhay' seed drill unit mounted on a metal frame which could be pushed by hand. At Marham, plots were 50 ft. long and 8.3 ft. wide in a 3 x 3 randomised block layout. Pelleted sugar-beet seed (cultivar 'Monotri') was sown at 3 in. spacing and 1 in. deep at three times: 12 April, 26 April and 10 May, using a 'Stanhay' drill tractor-mounted for the first sowing but mounted on the hand-propelled frame for later sowings. At Shouldham the number of seedlings emerged on 3 x 42 ft. of row per plot were counted on 23 May, 6 June and 20 July; at Marham similar counts on 3 x 43 ft. of row per plot were done on 17 May, 29 May and 6 June.

8.2.3. Results

The maximum number of seedlings that had reached at least the 2-4 rough-leaf (r.l.) stage from each sowing was used to calculate the seedling population. At Shouldham the first sowing had reached this stage on 6 June whilst the third sowing had not yet germinated. The seedling population for second to fourth sowings was calculated from the 20 July count when the first sowing had been singled. At Marham the 6 June count showed the first sowing at the 8-10 r.l. stage, the second sowing at the 4-6 r.l. stage and the third sowing at the 2-4- r.l. stage. The number of seedlings, expressed as a percentage of the total number sown, was calculated to represent seedling establishment from all sowings, whilst the seedling population was expressed in thousands per acre (Fig.42). At both sites the earliest sowing gave the most seedlings. At Shouldham significantly fewer ($P < 0.1\%$) seedlings established from the second, third and fourth sowings but at Marham differences were not significant. The particularly high seedling population at Shouldham was due to the particularly narrow (1.5 in.) spacing used whereas at Marham the seedlings were spaced at 3 in. Although no soil samples were taken on these experiments, observations confirmed that millipedes were aggregating around the seedlings and causing seedling loss.

8.2.4. DISCUSSION

Both at Shouldham and Marham soil cores, taken centred on seedlings on adjacent seed spacing experiments, indicated that numbers in the seedbed and the proportion of the millipedes that had moved into the rows increased from low numbers in April to high numbers in June. The successful establishment of the first sowing at Shouldham was therefore probably due to the presence of insufficient numbers of millipedes in the seedbed to cause a significant loss of seedlings or growth retardation. In contrast, the 12 May sowing may have coincided with a peak in millipede numbers and activity, resulting in severe damage

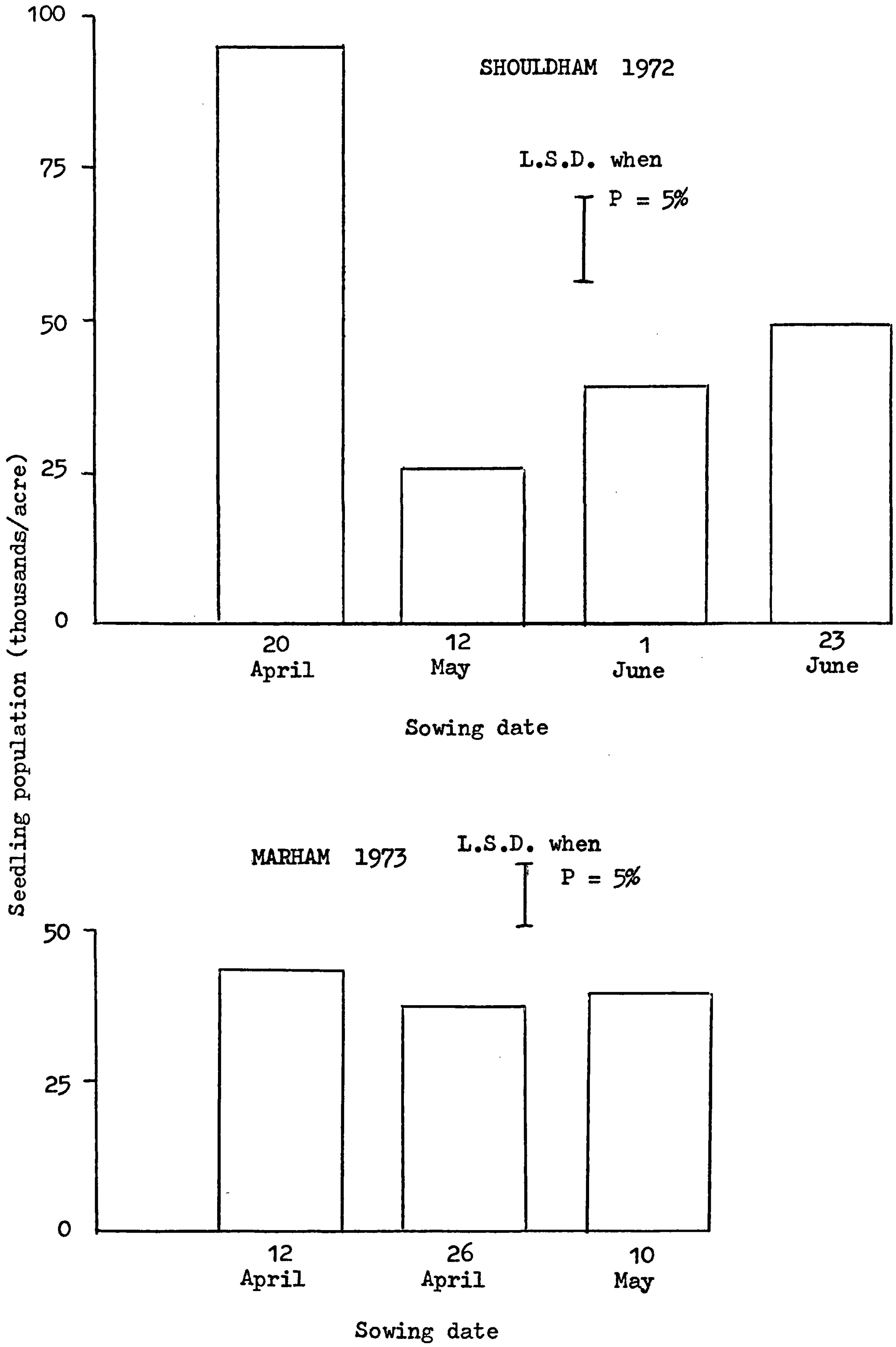


FIG. 42. EFFECT OF SOWING DATE ON SEEDLING POPULATION AT TWO SITES WHERE MILLIPEDES WERE PREVALENT.

and seedling loss. The apparent benefit of later sowings probably reflects both quicker emergence and a decrease in millipede numbers in the seedbed. At Marham, where the millipede population was both similar in size and age-structure to that at Shouldham, sowing at different times failed to give any significant benefits although a similar trend was evident. Other factors, linked with the experimental procedure, may also have determined whether one sowing would be more successful than another. Owing to the different sowing times, it was not possible that seedbed conditions could be uniform throughout all sowings. Any differences in level, firmness or compaction may influence the uniformity and percentage of seedling emergence (Hull & Jaggard, 1971) and although seed were drilled only when the seedbed was sufficiently moist and the same type of drill unit was used throughout, the initial drilling on the freshly cultivated seedbed would probably be the one most likely to produce the best seedling establishment.

8.3. EFFECT OF TEMPERATURE.

8.3.1. Introduction

Previous observations, both in the laboratory and in the field, suggest that sugar-beet seedlings can sometimes grow in the presence of millipedes without incurring damage. Since damage is the result of two interacting factors, millipede feeding activity and seedling vigour, then any factor which decreases the former and reinforces the latter will allow more seedlings to grow unharmed. Soil temperature is thought to be the main factor governing both millipede activity and rate of seedling growth; high temperatures ensure a high level of locomotory and feeding activity but rapid seed germination and establishment under these conditions may result in only minor damage being incurred. The timing of sowing, as demonstrated in a field experiment, may sometimes have a pronounced effect upon the number of seeds which develop into healthy seedlings. It is thought that, since soil temperatures are increasing during the spring, then seedling survival and extent of damage may be related to sowing time through the influence of soil temperature. A laboratory experiment was therefore carried out to investigate the effect of temperature on the extent to which sugar-beet seedlings become damaged by millipedes.

8.3.2. Materials and Methods

Rubbed and graded unpelleted sugar-beet seed (cultivar 'Amono') was left to germinate on moist filter paper within a closed plastic box in the laboratory. After three days those seeds which had begun to germinate were removed, the criterion being whether the radicle was just visible under the operculum. Ten seeds were carefully pressed into the surface of a 0.2 in. layer of moistened firm soil in each 3.5 in. diameter plastic Petri dish. Adult Boreoiulus tenuis millipedes, chosen at random from the culture kept at 5° - 10°C, were placed on the soil with the moistened tip of a fine paint brush. The soil in each dish was collected from the millipede's natural habitat and air-dried before use to kill any Collembola or mites that were present. For the first experiment fifty millipedes were placed in each of eight Petri dishes. Two Petri dishes A(i) and (ii), containing germinated seed and millipedes, were kept at each of the four temperature régimes; 4-5°C, 5-10°C, 15°C, 15-21°C, for a period of three days after which the seedlings were removed and examined for damage. Damage was assessed visually by a scoring method with a scale of 0-5 (0 = seedling free from damage, 5 = damage of sufficient severity to cause death of seedling). The total damage from each of the ten seeds of each replicate was recorded and ten fresh germinated seeds introduced into each Petri dish. After thirteen successive sowings at each temperature, over a two month period, the experiment was concluded and the percentage mortality of the millipedes was calculated.

For the replicates B(i),(ii) and C(i),(ii) a similar procedure was followed but thirty-five millipedes were used in each dish and a different temperature, 10-15°C, was substituted for the 15°C régime.

8.3.3. Results

The damage per seedling between each of the experimental series A, B, C, is not directly comparable since fewer millipedes were used in series A than in series B and C, and the experiments were conducted at different times: Series A from 22 August to 24 October, series B from 15 August to 12 October, series C from 9 October to 2 November 1972. For this reason the results from each series are shown separately in the figures.

Fig. 43 shows the mean damage score per seedling for each of the experimental series over a number of successive sowings (a range from 7 to 13) at five different temperature régimes: 4-5°C, 5-10°C, 12.5°C, 15°C and 15-21°C. At all temperatures, damage was usually most severe at the first sowing and there was a trend towards less damage at subsequent sowings. This effect was most marked when the germinated seed were kept at 15°C and especially at 15-21°C. For series B at 15-21°C, the seedlings could be grown free of damage after six sowings, after four sowings for C, but ten sowings for A. At 5-10°C, although the damage was variable and highest at the first sowing, it did not decrease to zero in series A, which was continued for thirteen sowings.

Fig. 44 shows the effect of the different temperatures on the total damage from all sowings. The results are a mean of damage scores from 130 seedlings from series A and 70 seedlings from series B and C, for each temperature. For each series the seedlings were damaged least at 4-5°C and at 15-21°C. Peak damage appeared to occur in the 5-10°C and 12.5°C régimes.

8.3.4. DISCUSSION

The distinct pattern of feeding behaviour between first and last sowings was shown at all the temperatures but there was a suggestion that at temperatures of more than 5°C feeding was proportionately

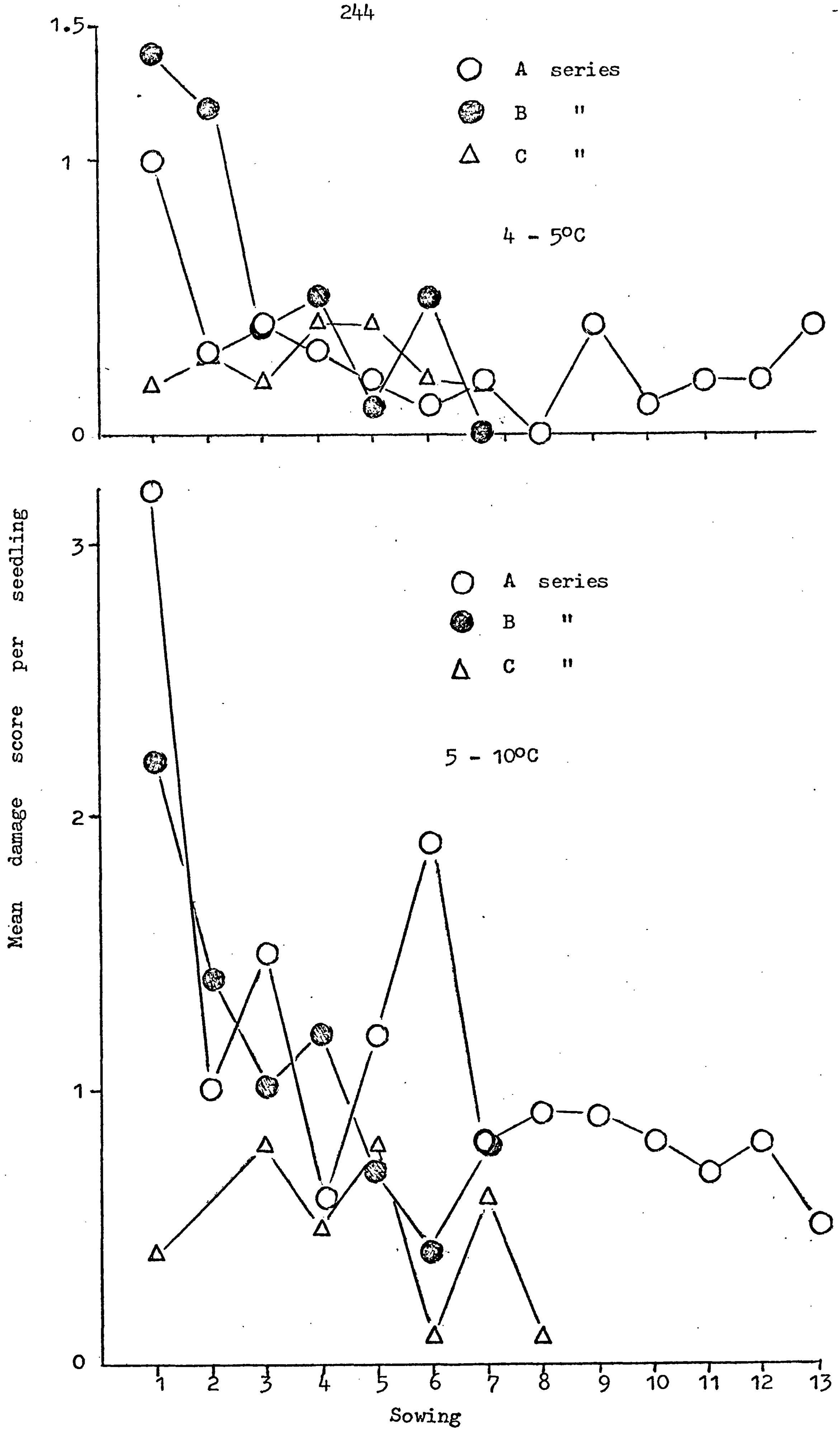


FIG. 43. <u>MILLIPEDE DAMAGE TO SUGAR-BEET SEEDLINGS AT SUCCESSIVE SOWINGS AT DIFFERENT TEMPERATURES</u>. continued on P. 245

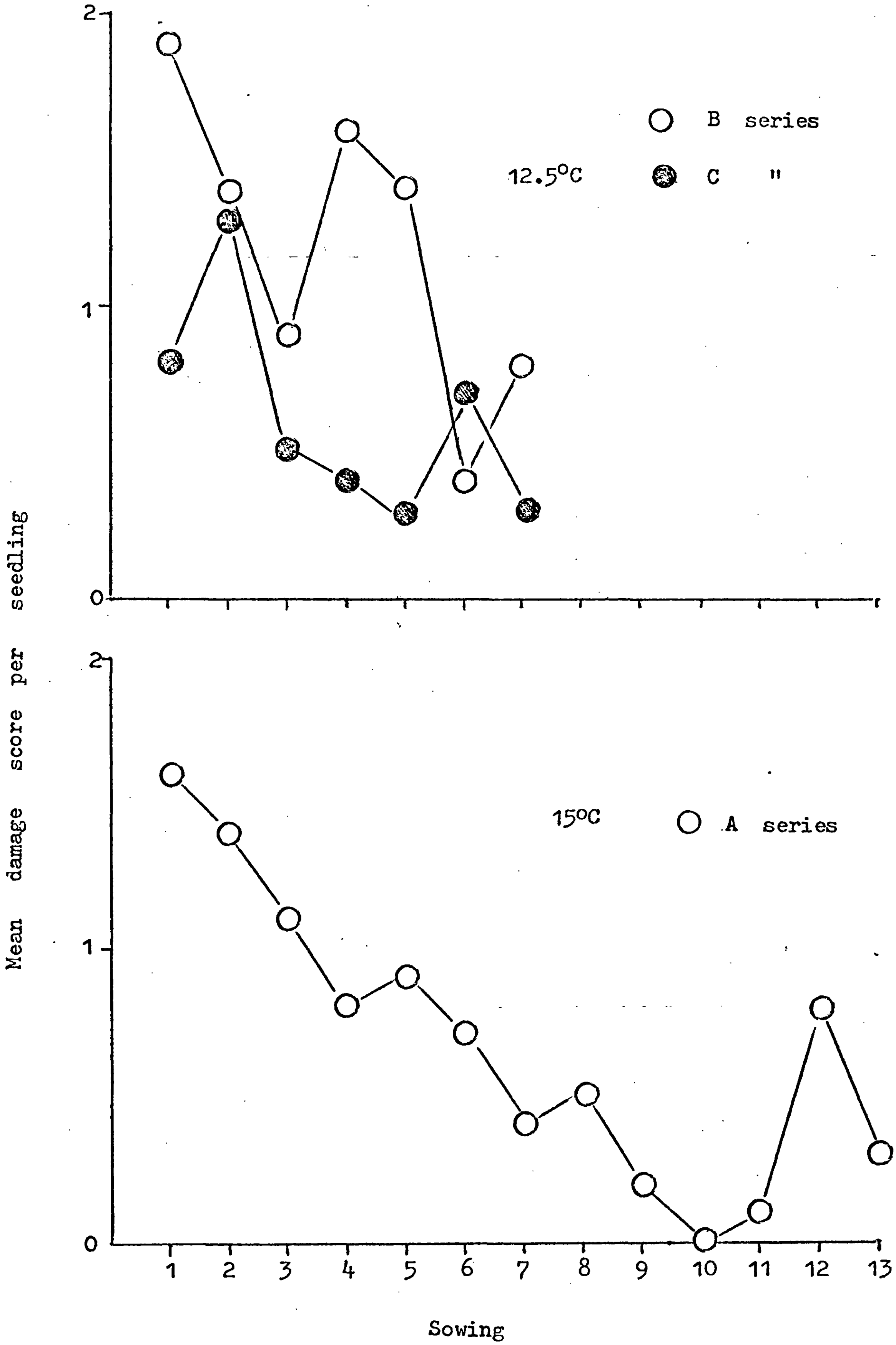


FIG. 43. (CONTINUED)

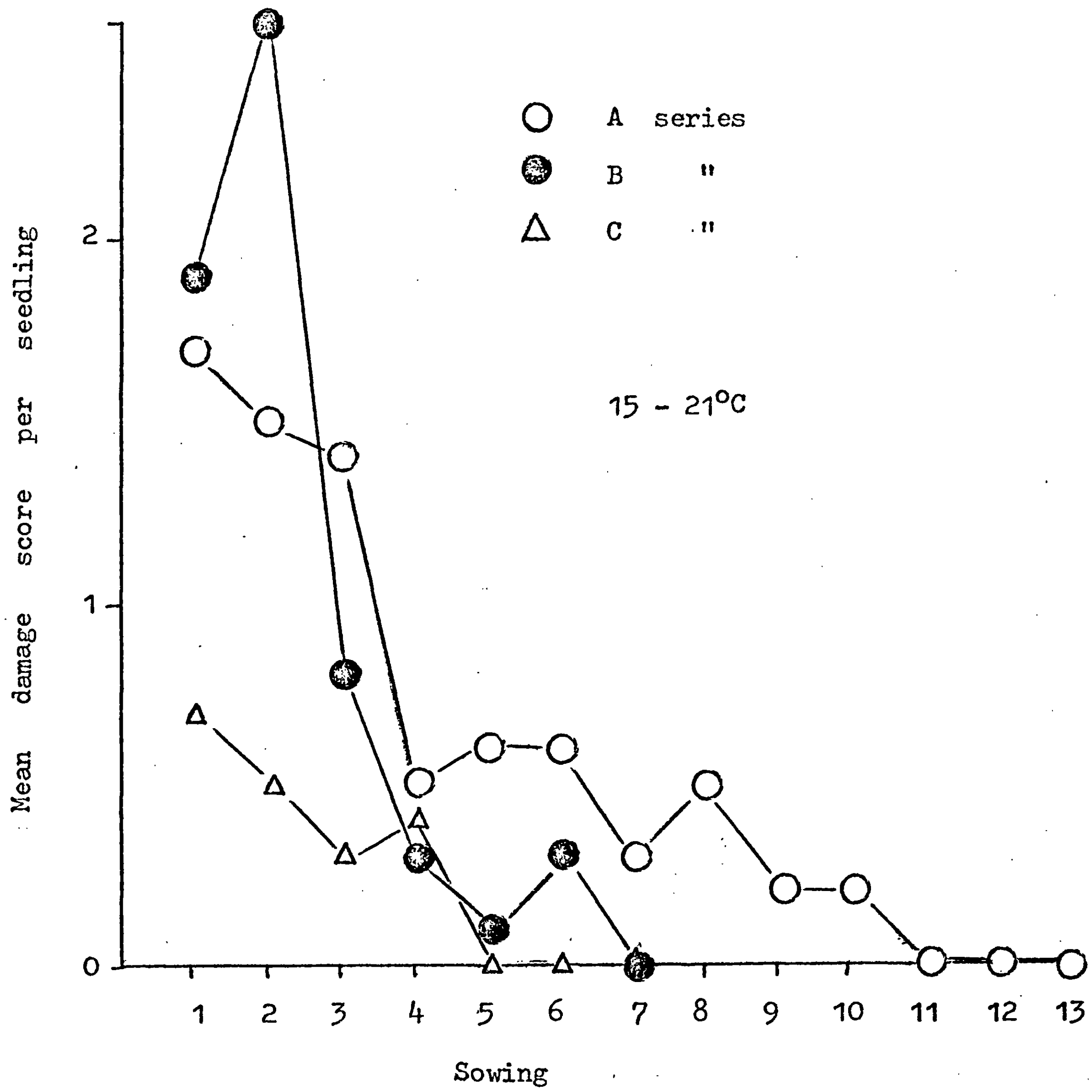


FIG. 43. (CONTINUED)

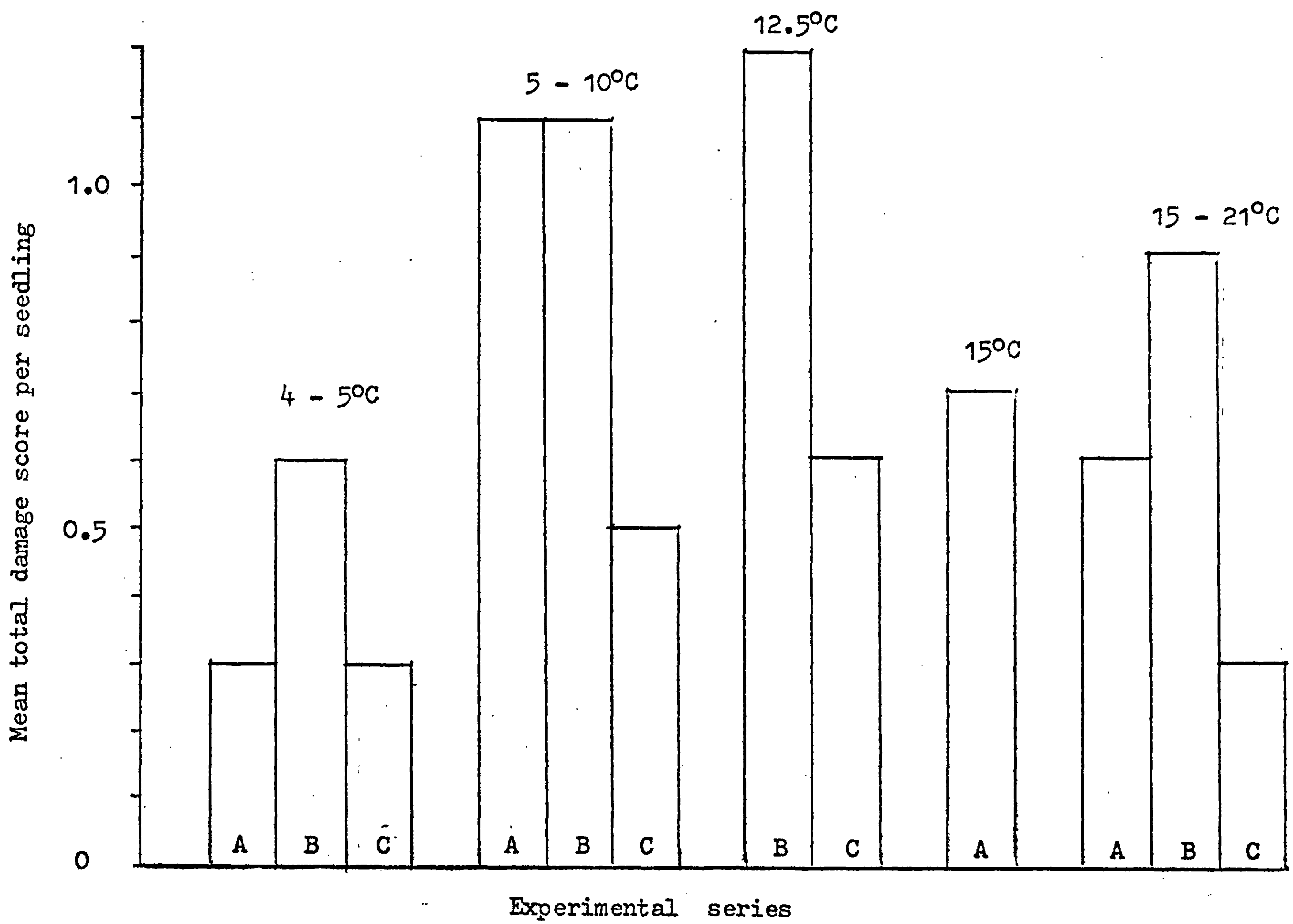


FIG. 44. EFFECT OF DIFFERENT TEMPERATURES ON THE TOTAL DAMAGE TO GERMINATED SUGAR-BEET SEED BY MILLIPEDES.

greater initially than at the final sowing. At the highest temperature feeding ceased after a number of sowings but was comparatively high initially. At 4-5°C, feeding, after two or three sowings, was consistently low. The general decrease in damage at subsequent sowings, especially at the higher temperatures, suggested that feeding activity may be followed by a non-feeding phase; observations of cultures indicate that this in turn is followed by a moulting phase. Michelbacher (1938) and Edwards (1961) found that the Symphylid Scutigerella immaculata exhibited a similar pattern in its feeding cycles. Since the histograms, showing total damage, are composed of the sum of the damage scores from every sowing, they reflect the temperature at which damage occurs at the most consistently high level throughout this period. That a temperature between 5° and 12.5°C seems to encourage most damage is in general agreement with the range of temperatures in the seedbed in April and May when most damage by millipedes occurs. The other factor operating, which may explain the differences in damage, is the effect of the different temperatures on the rate of growth of the seedlings. After three days the shoot length ranged from 0.1 in. at 4-5°C to 0.8 in. at 15-21°C. As the seedling shoots and roots increase in size the millipedes tend to cause less fresh damage, although they may still be active; a fresh sowing of germinated seed was often sufficient to cause a resumption of damage. At the lowest temperature, where the rate of seedling growth was slowest, the increased length of the period of maximum seedling susceptibility would be offset by the generally lower level of millipede activity.

8.4. EFFECT OF SOIL COMPACTION

8.4.1. Introduction

The amount of pore-space in a soil can govern both the amount of activity and species of non-burrowing soil invertebrates (Weis - Foch, 1948). The concept that soil compaction, by limiting the space between the soil crumbs, can be used as a control measure for the protection of crops against soil-inhabiting pests is not new. Ormerod (1890) proposed that it may be an effective measure against wireworms on the basis of some field observations and Petherbridge & Stapley (1935) observed that heavy rolling served as a useful measure of limiting loss of sugar-beet seedlings due to pygmy beetle.

The present studies tested the effect of 'loose' and 'compacted' seedbeds on numbers of the most common soil-inhabiting pests in the root zone of sugar-beet seedlings and recorded the effects on growth of seedlings not treated with any insecticide. A later experiment tested the combined effect of a soil-applied insecticide as an additional treatment.

8.4.2. Preliminary observations

The stimulus for studying the effects of soil compaction on soil-inhabiting pests originated from observations at Bottisham (Cambs.) in early June 1970 where the grower had reported that many seedlings were dying in a patch of ground at one side of the field. Closer inspection showed a regular pattern of alternate vigorous and poor growth running in parallel lines at 90° to the direction of drilling. Since the pattern of good growth was repeated at approximately 5 ft. intervals across the field and fitted the track dimensions of the tractor, it was thought that the effect could be due to soil compaction caused by the tractor wheels in earlier cultivations, prior to drilling the crop. An aerial photograph (plate 25) taken in August, when the foliage cover was at its maximum, showed clearly the striping effect across the field. The dark areas represent sugar-beet foliage whilst the white areas represent bare soil where the seedlings had died; many Boreoiulus tenuis and Blaniulus guttulatus millipedes were observed around the dying seedlings earlier, in June. The worst affected area, where observations were made, is enclosed within the white outlined area. Plate 26 is a photograph taken at 90° across the direction of the rows in late June, within the area marked in white in the previous plate, and shows clearly the pattern of crop growth; a pattern which was maintained until the crop was harvested. Fifteen soil cores, taken in both the areas of good and poor growth, showed that there were measurable differences in the degree of soil compaction. Mean bulk density values of 1.6 and 1.4 were obtained from cores 6 in. deep taken adjacent to seedlings where growth was good and where growth was poor, respectively. To test whether there were differences in the number of potentially injurious arthropods around the seedlings from the two areas, ten soil cores (2 in. diameter x 6 in. deep) were taken at random, centred on and including seedlings, from both areas in early June when most seedlings were at the 2-4 r.l. stage. The arthropods were extracted by crumbling the soil over cold water to which a little petroleum spirit had been added. Many soil-inhabiting pests were found in the root zone; the mean number per soil core from areas of compacted (good seedling growth) and loose (poor seedling growth) soil were respectively:-

PLATE 25. <u>AN AERIAL PHOTOGRAPH SHOWING THE EFFECT OF EARLY SEEDBED CULTIVATIONS ON THE PATTERN OF GROWTH OF SUGAR BEET IN A FIELD AT BOTTISHAM, 1970</u>.

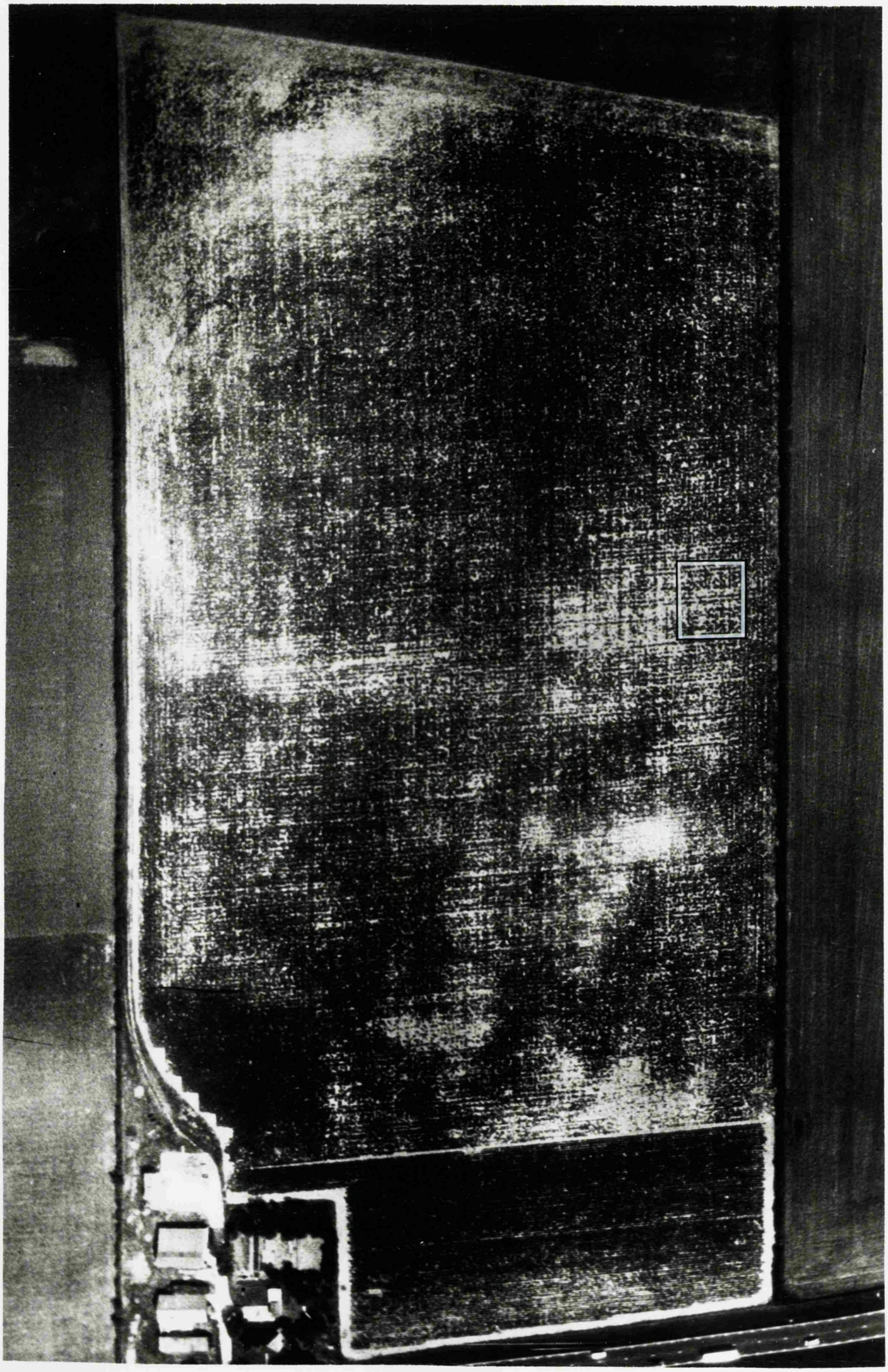


PLATE 26. THE DISTRIBUTION AND GROWTH OF SUGAR-BEET PLANTS IN A MILLIPEDE-INFESTED AREA WITH ALTERNATE STRIPS OF COMPACTED AND UNCOMPACTED SOIL AT BOTTISHAM, 1970.

Blaniulidae, 4.6 and 19.9; Brachydesmus superus, 0 and 0.5; Scutigerella immaculata, 0.3 and 2.1; wireworms, 0.1 and 0.2; pygmy beetle, 0.7 and 0.4.

It was postulated that the differences in seedling growth were due to differences in the degree of infestation by the different pests which were causing damage to the roots and impairing growth. Subsequent studies aimed at testing this hypothesis experimentally.

8.4.3. Materials and Methods

8.4.3.1 Sites and Experiments.

In 1971, at Bottisham, Welney (Norfolk) and Boston (Lincs.), randomised block experiments tested the effect of soil compaction on the distribution of soil-inhabiting pests and seedling growth.

A Stanhay drill sowed pelleted monogerm sugar-beet seed (cultivars 'Sharpe's Klein Monobeet' and 'Bush Mono') in 5-row plots. On the uncompacted plots the seedbed was kept as loose as possible and the inter-row spaces rotovated immediately after sowing; on the compacted plots, tractor wheels firmed every inter-row space and a garden roller firmed the seed rows.

In 1972 an improved technique was employed to obtain a higher degree of compaction at a site at Shouldham (Norfolk). The experimental area was split longitudinally into two areas each 12 ft. 6 in. wide (1.5 drill widths). The soil in the whole of the left-hand area was compacted by running a tractor up and down the eight 50 ft. long plots. The right-hand area was harrowed and left uncompacted. The whole area, 400 ft. long x 25 ft. (3 drill widths) wide was drilled with pelleted sugar-beet seed (cultivar 'Amono'), not treated with an insecticide, at 4.5 in. spacing with plots 5 rows wide (6 ft. 8 in.). Half the plots in each block were treated with oxamyl insecticide applied from a Horstine Farmery 'Microband' granule applicator mounted on the drill, at a rate equivalent to 16 oz. a.i./acre.

8.4.3.2. Sampling and extraction of soil-inhabiting pests.

Some details of soil sampling and extraction at experimental sites are shown in Table 34. Each soil core was taken centred over and including a seedling (in the late cotyledon stage) and placed in individual aluminium screw-top cans. In the laboratory the creatures were extracted by the flotation technique using petroleum-spirit. At Shouldham, where the seedlings were damaged by millipedes Boreoiulus tenuis and Blaniulus guttulatus, no soil samples were taken on the experiment.

8.4.3.3. Soil density measurements.

Soil density gives a measure of the degree of soil compaction. The sampling unit was a core 3.9 in. diameter x 2.3 in. deep at all sites, except Shouldham where the auger cut a 1 in. x 1.6 in. deep core. Soil cores were taken on each plot, both in the row (between the seedlings) and midway between the rows on both compacted and uncompacted plots and placed in sealed aluminium cans. In the laboratory the soil was dried in a ventilated oven at 105°C for 24 hrs. and weighed. The degree of compaction was expressed as a bulk density value in gms. per cc.

TABLE 34. SOIL SAMPLING AND EXTRACTION OF PESTS; FIELD EXPERIMENTS 1971-1972.

	Site	Sample unit cores	Units/ treatment	Treatments	Pests extracted	Condition of extract
1971	Boston	3.9 x 2.3 in.	24	Compacted v.loose seedbed	Pygmy bettle	dead
	Bottisham	" "	8	"	Millipedes	dead
	Welney	" "	12	"	Pygmy beetle	dead
1972	Shouldham	none	none	Compacted v.loose seedbed ± insecticide	Seedling counts only	

8.4.3.4. Seedling observations.

Seedling root damage was assessed at the Boston site on 3rd June 1971 where pygmy beetle were causing damage. Twenty seedlings were removed at random from each plot, roots washed and both the number of bites on the roots counted and a damage score (0 - 4) given to each seedling root (0 = no damage, 4 = severe damage).

Seedlings were counted on a measured length of row on the three centre rows of each plot at every site two or three times until the maximum number of seedlings had emerged.

8.4.4. Results

8.4.4.1 1971 Experiments

All data was analysed by Analysis of Variance. Few millipedes were found in the cores from Bottisham and although more were present in cores from uncompacted plots the difference was insignificant. More pygmy beetle were in cores from compacted plots at Boston and Welney where the infestation was sufficiently large to cause considerable seedling loss but differences at both sites were not quite significant at the 5% level of probability (Table 35).

Seedling damage, assessed at Boston only, indicated that seedling roots from uncompacted plots had significantly more damage than from compacted plots; the number of individual bites per seedling were also greater but not significantly so. Significantly more seedlings established on compacted plots at Boston and Welney sites, both of which had pygmy beetle infestations. At Bottisham, where seedlings were damaged by millipedes, there was no difference in seedling establishment but some losses were attributable to field-mice (Table 36).

8.4.4.2. 1972 Experiment

The results of this experiment were also analysed by Analysis of Variance but physical and chemical treatment effects were separated to evaluate their individual contribution (Table 37). Significantly more seedlings established on uncompacted plots and where the soil had been treated with oxamyl than on compacted and untreated plots. Most seedlings were recorded on plots which were uncompacted and which additionally had received insecticide (Table 36).

TABLE 35. THE EFFECT OF SOIL COMPACTION ON THE NUMBER OF PESTS IN THE ROOT ZONE OF SUGAR-BEET SEEDLINGS.

Mean number of pests per sample unit

Site: Pest:	Boston 1971 Pygmy beetle	Bottisham 1971 Millipedes	Welney 1971 Pygmy beetle
Treatment			
Soil uncompacted	4.25	1.75	6.60
Soil compacted	5.25	0.75	6.80
S.E.D. ±	1.25	1.59	0.21
Degrees of freedom	15	9	15

TABLE 36. SEEDLING OBSERVATIONS.

	Seedlings per yard of row				Seedling damage Boston, 1971.	
Site Treatment	Boston 1971	Bottisham 1971	Welney 1971	Shouldham 1972	No. of beetle bites/ seedling	Mean score /seedling (0-4)
Soil uncompacted	3.71	7.47	0.60	4.86	4.67	1.32*
Soil compacted	4.51*	7.50	1.30**	4.10	4.40	1.20
Soil loose +insecticide	-	-	-	6.12	-	-
Soil compacted + insecticide	-	-	-	5.28	-	-
S.E.D. ±	0.25	0.41	0.21	-	0.35	0.07
Degrees of freedom	15	9	15	-	15	15

*, **, denotes differences significant at P <5% and 1% respectively.

TABLE 37. COMPACTION AND INSECTICIDE TREATMENT EFFECTS ON SEEDLING NUMBERS; SHOULDHAM, 1972.

Treatment / Observation	Physical treatment: Uncompacted soil	Physical treatment: Compacted soil	Chemical treatment: Untreated	Chemical treatment: Oxamyl
No. of seedlings (thou./ac.)	47.8*	40.8	39.0	49.6**
S.E.D. ±	2.3		2.3	
Degrees of freedom	9		9	

*, **, denotes differences significant at P <5%, 1% respectively.

TABLE 38. SOIL COMPACTION VALUES AT EXPERIMENTAL SITES IN 1971 and 1972.

Mean bulk density (gms./cc.) per sample unit (IR = in row (BR = between rows

	Boston 1971 IR	Boston 1971 BR	Bottisham 1971 IR	Bottisham 1971 BR	Welney 1971 IR	Welney 1971 BR	Shouldham 1972 IR	Shouldham 1972 BR
Uncompacted	1.4	1.3	1.2	1.1	1.3	1.2	1.3	1.3
Compacted	1.5	1.5	1.4	1.4	1.4	1.4	1.4	1.4

8.4.4.3. Compaction measurement

Compaction was expressed as bulk-density (gms./cc.). Table 38 shows a comparison of 'in' and 'between' row values for both uncompacted and compacted plots.

Bulk-density values were always greater on compacted plots by up to 14% but the between-row spaces were usually less compacted on 'uncompacted' plots than in the rows.

8.4.5. <u>Discussion</u>

In the field experiments compacting the soil by using the tractor wheels benefitted seedling growth only where pygmy beetle were causing damage. At the two sites having millipedes seedling growth was often significantly worse on compacted plots; this is contrary to what was expected from preliminary observations. The failure to reproduce the benefits of compacting the soil at millipede-infested sites could be due to many factors: that the degree of compaction was insufficient, that it did not extend deep enough into the sub-soil, that the timing of the compaction may be important or that the damage was not sufficiently severe to outweigh any normal adverse affect on seedling growth. The first point may be valid, since the bulk-density values recorded in 1970, at the site where the preliminary observations were made, were higher than achieved artificially on experimental plots. The depth to which the compacted zone extended was not measured. The compacted strips seen at Bottisham in 1970 were probably caused during seedbed preparation, perhaps some weeks before drilling, where the tractor was following previous tracks;

this may have been before the millipedes had moved up from the sub-soil into the seedbed. The subsequent resistance to free movement produced by the compacted soil may have caused a redistribution at depth or largely prevented millipedes coming up to the seedling root-zone on the compacted strips. In the 1971 experiment at Bottisham, on the same type of soil, some slight reduction in millipede numbers were induced on the compacted plots but the variation was too great within the sample for any true treatment difference to assume significance; the amount of damage caused by the millipedes was less than in 1970.

Although pygmy beetle numbers per seedling root zone at the two sites in 1971 were higher on the compacted plots, damage to roots and seedling numbers were greater on the uncompacted plots. The obvious conclusion is that the more compacted soil was restricting their mobility to move from one seedling to the next where they would otherwise have caused fresh damage. Pygmy beetle may be more affected by the usual amounts of soil compaction than millipedes because they are not adapted for moving extensively within the soil and must rely more on existance of fissures; pygmy beetle can readily be observed walking across the soil surface in infested fields. It is particularly easy for them to enter the soil where a seedling is growing because the soil displacement, caused by the extending seedling hypocotyl, creates a fissue around the seedling, particularly if the soil is relatively dry. A heavy rain may help to seal the surface around seedlings but no information is available on the effects of moisture on numbers of beetles in the root zone of seedlings and it is hypothetical that rain may reduce damage by closing the fissures by which the beetles would normally enter the soil.

Other workers studying soil-inhabiting pests have recorded the beneficial effects of compaction: Hunter (1967) found that cultivation, followed by compaction, could eradicate slug populations and noted the beneficial effects of tractor-wheelings on the growth of a wheat crop. Michelbacher (1938) had observed that rows of sugar beet that had been crossed by a tractor were more vigorous than the rest of the crop where Symphyla were causing damage. Gough & Edwards (1971) reported the beneficial effects of soil compaction against Symphyla attack of corn seedlings whilst Heijbroek (1972) has noted that cases of serious damage by Collembola (Onychiurus spp.) were rare on compacted soils. There are thus many observations which support the concept of soil compaction as a viable method of reducing damage to arable crops where a soil-inhabiting pest is present.

We must now consider the effects of compaction at sites where pests are absent if it is to be considered as a preventative treatment.

Agronomic studies have shown that compacting a seedbed to a bulk density value of 1.65 gm/cc. gives a poorer seedling establishment than where the soil was only 1.47 gm./cc. (normal seedbed) or 1.30 gm./cc. ('loose' seedbed), (Jaggard, 1971, 1972). The values recorded on compacted plots in the present study where pests were present relate more closely to this 'normal' value and would therefore not be especially detrimental to growth if pests had not been present. Clearly in any further studies a higher degree of compaction on 'compacted' plots should be attempted. The reason for the low levels of compaction could have been due to the dry soil conditions at the time of drilling in the experiments.

The experiment at Shouldham in 1972 demonstrated the effectiveness of oxamyl in protecting seedlings against millipedes, particularly if the soil was uncompacted (at this site the soil on uncompacted plots was not loosened with the rotovator). The apparent increase in effectiveness of the insecticide in these conditions was possibly attributable to increased ease of access of millipedes to the chemical, though no soil samples were taken to lend support to this conclusion.

8.5 EFFECT OF AN ALTERNATIVE FOOD

8.5.1 Introduction

The remains of previous crops, such as cereals or potatoes, can often be found in a field which has been sown with sugar-beet seed. It is common to find millipedes, which are polyphagous, accumulated around the rotting remains of these crops which serve as food, shelter and perhaps a breeding site. Although these materials eventually decompose during the course of a normal summer millipedes can be found aggregating around them when they are adjacent and close to germinating sugar-beet seedlings to which they are also attracted. In order to establish whether this alternative food material is sufficiently attractive to arrest the normal food-searching activity of millipedes an experiment in the laboratory, with two common species of millipedes, tested whether there was any effect on the extent of damage to sugar-beet seedlings when they were grown in soil to which a rotted cereal straw/manure mixture had been added.

8.5.2 Materials and Methods

Both Brachydesmus superus and Boreoiulus tenuis have been found in close association with organic material in the soil, the latter species was particularly numerous at a site at Shouldham (Norfolk) in 1972; from here, samples of both millipedes and organic material were collected in June. The organic material was composed of cereal straw mixed with pig manure which had been applied to the soil in the previous winter and had partially decayed. The soil, a peaty loam, was oven-dried, then remoistened to approximately 15-20% m.c. The cereal straw mixture was shredded and thoroughly mixed with a sample of soil. Sixteen glass Petri dishes were prepared, eight with a 0.4 in. layer of soil only (control) and eight with the straw mixture. Into each of four containing each substrate were placed thirty B. superus (stadia VI and VII in equal numbers) and four with 20 adult B. tenuis. Five raw sugar-beet seed (cultivar 'Amono') which had been pre-germinated, were placed on the surface of the soil in each Petri dish. Butt-jointing lids were fitted, taped over to provide a sealed chamber, and the Petri dishes were left in an unheated room where the temperature varied from 6 to 12°C. The seedlings were removed after four days when the radicles were about 0.8 in. long and scored for damage on a 0-5 scale as in previous experiments. Five fresh pre-germinated seeds were introduced into each Petri dish and the experiment repeated for a total of nine sowings with B. superus and 13 sowings with B. tenuis.

8.5.3 Results

The total damage scores for each sowing (four replicates each of five seed) were calculated for the control and where straw had been added. The mean damage score per sowing for the two treatments was compared by a 't' test for each millipede species and shown in Fig.45. The mean damage for each sowing was greater for those seedlings attacked by B. tenuis than for B. superus and with each millipede the seedlings grown in the soil/straw mixture had significantly less damage compared with those grown in the untreated 'control' soil. The respective values of 't' for B. superus and B. tenuis were t = 3.7 (P <1%)

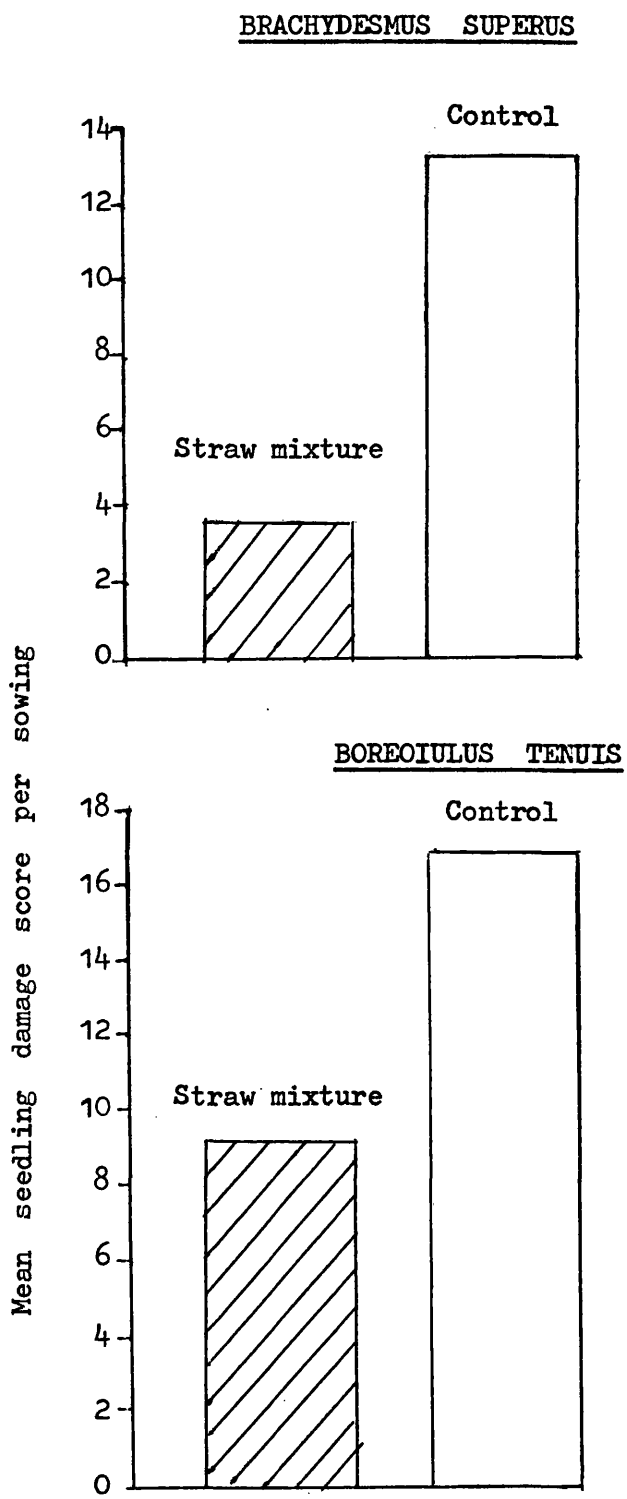


FIG. 45. EFFECT ON DAMAGE BY MILLIPEDES OF GROWING SUGAR-BEET SEEDLINGS IN A SOIL/STRAW MIXTURE.

and t = 7.3 (P <0.1%) with 10 and 32 degrees of freedom for each species respectively.

8.5.4 Discussion

The total number of sowings for seedlings in Petri dishes with each millipede species reflected the total period over which they continued to cause damage. For B. superus the ninth sowing was undamaged after four days, whilst B. tenuis continued to damage seedlings at the thirteenth sowing. It could be argued that the reduction in damage to seedlings grown on the soil/straw mixture was due to the reduction in millipede activity because the straw may have provided a more favourable resting place, but care was taken in the preparation of the mixture to obtain an overall texture similar to that of soil itself by shredding the straw fine.ly In previous studies millipedes have been fed on a variety of organic material: senescent sugar-beet leaves, boiled barley and Tick-bean, agar and maize; all have been attractive before the advent of fungal growth which can affect their chemical composition and odour. In the field there is much evidence of millipedes feeding on straw buried in the soil and they undoubtedly contribute towards its breakdown; the fungal flora of straw may also be not unattractive. A laboratory study tested the amounts ingested of four different substrates of approximately equal moisture: straw/soil/manure mixture, loamy peat soil, potting compost, loam soil, by adult B. superus in Petri dishes. The number of faecal pellets that were deposited on radially arranged filter paper squares lying on the substrate surface were counted. The millipedes were remixed and allocated to a different Petri dish after each observation to allow for differences in batches. After six separate 24 hr. counts most pellets (835) were produced on the straw/soil/manure mixture and least (241) on the loam, showing that the former was a particularly attractive food. The filter paper squares which had been soaked in various solutions, including sugar, were also eaten and when sufficiently thin area loss was assessed. Whereas it was almost completely eaten on the loam and peaty loam substrates, almost none of the filter paper on the straw/soil/manure substrate was consumed, suggesting that this substrate per se was an attractive alternative food source. The attraction of this mixture as a habitat was also confirmed in choice experiments where the numbers of millipedes were counted within the substrate at intervals.

Although the straw mixture may attract and encourage millipedes to feed on it and so divert millipedes from attacking seedlings, it can have an adverse effect in the long term by encouraging breeding and therefore result in the build-up of a larger population (Morris, 1927); field observations in the present study confirm that it provides a suitable substrate for the youngest stadia.

9. CONTROL OF SOIL-INHABITING PESTS OF SUGAR-BEET SEEDLINGS BY INSECTICIDES

9.1. INTRODUCTION

All commercial sugar-beet seed is treated with a dressing of dieldrin (0.2%, w/w), an insecticide particularly effective against wireworm (Dunning and Winder, 1966). Wireworms were the most damaging soil-inhabiting pest from 1947-1956 (Dunning, 1975). Although wireworm incidence has declined markedly in recent years seedling losses due to soil-inhabiting pests still remain high because other pests such as millipedes and pygmy beetle now cause more damage. In recent years Government bodies responsible for pesticide usage have stimulated the search for a replacement non-organochlorine compound which will protect sugar-beet seedling roots against current pests. Two methods of protection are being sought - to find a non-persistent wide spectrum insecticide for incorporation into the seed pellet and to find a suitable formulation for seed-furrow treatment. Methiocarb is currently proving to be the most promising insecticide and its performance was compared to other compounds in both seed and furrow formulations. Different rates were tested to find a concentration that offered optimum protection from pest attack, whilst remaining non-phytotoxic to the seedling. Although most fields do not have pest problems, seed treatment is an insurance against cases of mild attack which are the most numerous. In specific cases, where fields have had a history of pest attack and where a more widespread treatment is required to ensure seedling survival, the current recommended insecticide is Gamma-BHC, which, although of less persistence than other organochlorine compounds, is also thought to be a potential hazard for wild-life and for which a replacement is also required.

The aim of the present study was to show how the insecticides, currently being tested either as seed, furrow, or as an overall spray, affect the numbers of the most common soil-inhabiting pests in the root zone of sugar-beet seedlings and, where possible, estimate their comparative activity in killing or repelling the pests and providing protection to the seedling. Seedlings suffer by incurring feeding lesions on their hypocotyl and root system which can result in restricted growth or even death. Assessment of the degree of protection afforded by the insecticide was by sampling the soil around and including seedling root zones and assessing performance by comparing seedling establishment (the percentage of seedlings emerged from seed sown), the main criterion for assessing insecticide performance in the field.

Although the study was mainly concentrated on soil-inhabiting pests which were common at the experimental sites, the effect of some insecticides on Acarina (mites) was noted in some later experiments since they have been shown to be very sensitive to pesticide residues (Edwards, 1965).

Studies in the laboratory tested some insecticides in the pellet against millipedes.

9.2. MATERIALS AND METHODS

9.2.1. Sites and Experiments

In the four year study experiments were carried out at chosen sites thoughout East Anglia; sites were often selected for one particular pest and one or more experiments at a site tested a range of compounds in a commercial crop; (Plate 4 shows the geographical location of sites referred to in the present study). The experiments were of two types, according to the formulation of the insecticide used -: seed treatment and furrow treatment. These experiments are described separately.

In a 'seed treatment' field experiment small quantities of each insecticide (up to 0.8% by weight) were incorporated around the seed (technically a fruit enclosing the seed) during the pelleting process where the 'seed' are made spherical by covering with a clay mixture. Although each experiment often compared more than 20 treatments only a few were selected for sampling. These were usually the untreated control, dieldrin and methiocarb seed treatments but sometimes gamma-BHC, mecarphon or PP505. In most seed-treatment experiments, except at Stalham, the normal recommendation of gamma-BHC was included as a direct comparison, this was applied either as an overall spray at 16 oz. ai./ac. on bare soil before drilling followed by harrowing to incorporate with the top 6-9 in. or, alternatively, as a treatment in the seed-furrow when conditions prohibited spraying.

The seed-furrow treatments, either as a granule or liquid formulation, were applied at the time of drilling directly into the furrow where pelleted seed, untreated with other insecticide, was sown. Liquids were applied by a simple gravity-flow system with a 9 ft. head and granules by a Horstine Farmery 'microband' granule applicator mounted on the drill. Both liquids and granules were placed in the bottom of a V-shaped furrow immediately after seed placement. The following seed-furrow applied treatments were tested: gamma-BHC and aldicarb both at 4 or 16 oz. ai./ac. and oxamyl at 4, 8, or 16 oz. ai./ac. Martin (1971) describes all of the compounds tested except PP505 (oxime carbonate compound).

Experiments were of a completely randomised block design with plots usually five rows wide and between 48 ft. and 70 ft. long. The seed was drilled at 3 in. spacing and between 0.8 in. and 1 in. deep on all experiments; rows were 20 in. apart. The seed-furrow treatments were applied to single rows only, separated by a discard row. At Stalham single row plots were used for the seed-treatment comparison but five row plots where overall sprays were used. Experiments at Welney, Boston, Benwick and Terrington were drilled with beet for the second consecutive year on fields where the previous beet crop had been damaged by pygmy beetle; the objective was to increase the probability that the new crop would be damaged by beetles overwintering in the soil. Others, Shouldham and Stalham, had a high population of millipedes *Boreoiulus tenuis* and Collembola (*Onychiurus* spp.) respectively. At Littelport, pygmy beetle, Collembola and Symphyla (*Scutigerella immaculata*) were present in the soil while at Cottenham, the pest was wireworms (*Agriotes* spp.) and at Gedney, millipedes (*Blaniulus guttulatus*).

9.2.2. Sampling and Extraction of Pests

Details of the sampling are shown in table 39. Single core sample units of 2.3 in. diam. x 3.9 in. deep were used in 1971 and 1972 only. In 1973 and 1974 bulked samples each comprising 8 (1 in. diam., 3.9 in. deep) cores were taken from each plot. Cores were taken at random in the row centred over and including a seedling on seed treatments at Boston, Welney, Littleport, Terrington; furrow treatments at Boston, Benwick and Cottenham and gamma-BHC spray treatments at Boston, Welney, Shouldham. At Stalham in 1974 the sample was composed of 1 yard lengths of row on seed treatments and gamma-BHC sprayed plots, whilst at Gedney plant samples, only, were taken on gamma-BHC spray and oxamyl spray treated plots.

9.2.2.1. 1971 Experiments

Soil cores were taken on the following treatments: control, dieldrin,0.2%; methiocarb,0.8%; gamma-BHC,0.2% pelleted seed treatments and gamma-BHC sprayed plots. In the laboratory the arthropods from soil samples were extracted from the soil by the petroleum-spirit flotation method described in section 3.3.1. but those previously either live or dead could not be differentiated. At Boston a similar sampling and extraction method was employed on the furrow-applied treatments but the pygmy beetle were separated into 'whole beetles', which were undamaged and were either previously live or recently dead prior to extraction and 'sections of beetles' where either heads, thoraxes or elytra were recorded; these beetles were probably dead before extraction.

TABLE 39 SOIL SAMPLING OF FIELD EXPERIMENTS WHERE INSECTICIDES WERE TESTED

Year	Site	Placement of insecticide	Sample unit size (diam.x depth)	Number of sample units/ treatment	Arthropods extracted	Condition of arthropods after extraction
1971	Boston	seed and spray	1 core 2.3 x 1.9 in.	12	C,M,Mi,P	dead
	"	furrow	"	16	C,M,P	"
	Welney	seed and spray	"	8	C,M,P	"
1972	Benwick	furrow	1 core 2.3 x 1.9 in.	9	C,Mi,P	dead (P=live + dead)
	Shouldham	spray	"	12	C,M,Mi	live
1973	Littleport	seed and spray	8 cores, each 1.0 x 3.9 in.	5	C,P,S	live + dead
	Terrington	seed and furrow		4	P	" "
1974	Cottenham	furrow	8 cores, each 1.0 x 3.9 in.	5	W	dead
	Stalham	seed and spray	1 yd. length of row	12	C	live + dead
	Stalham	spray	" " "	16	C,Mi	" "

Key : C = Collembola, M = millipedes, Mi = mites, P = pygmy beetle, S = Symphyla, W = wireworms

9.2.2.2. 1972 Experiments

At Benwick the numbers of both live and dead beetles were recovered from 9 cores taken at random from each treatment: control, gamma-BHC (10 oz. ai./ac.), oxamyl and carbofuran (both 10 oz. ai./ac.). Owing to the lack of healthy plants on the control plots, cores, usually centred over a healthy seedling, were taken over dying plants or in gaps in the row. The soil cores were first crumbled over cold water and the live and dead pygmy beetles picked off the surface of the water with a fine brush, petroleum-spirit was added and the soil stirred, any remaining live beetles would now have been killed and these were filtered off in the usual way. An estimate of the probable number of living and dead beetles in this second extraction was obtained from the respective ratios obtained from the first extraction using water only. The total number of living and dead beetles was calculated from the estimates in both extractions. In assessing the total live and dead beetles in each core it was assumed that the initial extraction, the water flotation, did not favour the recovery of either live or dead beetles. Onychiurid Collembola and soil mites were also recorded from the petroleum-spirit stage but could not be separated into live and dead.

At Shouldham twelve cores centred on a seedling at random were taken on untreated plots and those treated with gamma-BHC spray. Each sample unit was divided into two 5 cm. deep units and the arthropods extracted on a high-gradient cannister apparatus; details of its operation are given in section 3.3.4.

9.2.2.3. 1973 Experiments

Seed, furrow and overall spray treatments were sampled at both Littleport and Terrington. Although only 4 or 5 samples were taken per treatment, the new sampling technique, using bulked cores, meant that this represented between 32 and 40 seedlings per treatment. Collembola, pygmy beetle and Symphyla were extracted by crumbling the soil over the surface of cold water and live and dead individuals were separated.

9.2.2.4. 1974 Experiments

At Stalham, a single row plot, randomized block experiment compared the following treatments: untreated seed (control); dieldrin, 0.2%, and methiocarb, 0.8%. Seedlings were sampled on April 29 when 1 yard of row was removed at random with a trowel to a depth of 3 in. from each plot; this approximated to 2 litres of soil weighing between 5 and 7 lbs. In the laboratory the seedlings were picked out, washed and scored for root damage. This was inevitably a subjective measurement and depended both on the severity of damage and number of holes. A score on a 0-5 scale was given for the damage on each seedling root and between 10 and 13 seedlings were recovered from each plot. Collembola were extracted by crumbling the soil over a vessel filled with cold water; they were picked off the surface with the tip of a fine paint brush and placed on black filter paper in a Petri dish where the number of living and dead individuals was counted. A 'live' classification applied only to those individuals which were capable of locomotory movement. Those classed as 'dead' showed either no movement or spasmodic movement of the body when touched with a paint brush.

Also at Stalham, plots sprayed with gamma-BHC at 16 oz. ai./ac. and control plots (unsprayed) were sampled on 6 April when most cotyledons had risen clear of the soil surface. Soil was removed from four (1 yd.) lengths of row in each five row plot as in the seed treatment experiment. The seedlings were washed, scored for damage on a 0-5 scale and living and dead Collembola and mites separated out as previously described.

At Gedney the sugar-beet seedlings did not suffer from pest damage at the usual time due to drought conditions in April and May; millipedes, B.guttulatus appeared in the root zone following the first period of wet weather, in June. On 13 June, shortly after singling, 10 plants were removed at random from control (untreated) plots and plots sprayed with gamma-BHC (16 oz. ai./ac.) and oxamyl (8 oz. ai./ac). The roots were scored for millipede damage on a 0-5 scale in the laboratory.

At Cottenham, soil samples were taken at random from each of the following treatments: control, oxamyl and aldicarb (both at 4 and 16 oz.). The wireworms were extracted using petroleum-spirit and the seedlings removed and scored for root damage on the usual 0-5 scale.

At all sites seedlings were counted on fixed length of row on each plot immediately before singling. Seedling establishement was calculated from the known number of seeds sown.

9.2.3. Laboratory tests with millipedes.

9.2.3.1. Effect of some pelleted seed treatments on seedling damage and millipede mortality

Some preliminary experiments on the effects of insecticides incorporated into the seed pellet on damage to seedlings and millipede mortality were carried out using adult *Brachydesmus superus*. The following treatments were tested: - control (untreated); dieldrin, 0.2% and 0.8%; methiocarb 0.2% and 0.8%; carbaryl 0.8%. In the first preliminary experiment 5 seed and 10 millipedes were placed upon a layer of moist, peaty, soil in a crystalizing dish which was fitted with a polythene cover retained by elastic bands; two dishes were used per treatment and they were kept in darkness at 6-7°C. After 17 days the number of dead millipedes and the number of germinated seedlings that had been damaged were recorded.

The second experiment used *Blaniulus guttulatus* with pelleted seed of the following treatments: - control; dieldrin, 0.2%; methiocarb and mecarphon, both at 0.2%, 0.4% and 0.8% and PP505, 0.8%. Air dried sandy loam soil from a millipede site was packed loosely into square plastic pots (3.6 in. square x 2.4 in. deep) each having four drain holes and lined with a thick moist pad of filter paper. Each pot was placed in groups of six in a tray containing wet sand which served as a water reservoir. Five pelleted sugar-beet seed were placed in each pot in moistened soil at a depth of 0.5 in; five replicates were prepared for each of the nine seed treatments. The pots were left for 3 days in an unheated room at 15°- 17°C to initiate germination. 15 adult *B. guttulatus* were placed in each of the 45 pots which were then left in a controlled environment growth room set to simulate spring climatic conditions: light (0600-2000 hrs.); temperature, 6°C for 6 hrs (2401-0601 hrs), 12°C for 18 hrs (0600-2400 hrs).

After 4 weeks, when the seedlings had reached the late cotyledon stage, the number of seedlings that had germinated were counted, the root damage scored on a 0-5 scale and millipede mortality determined for each pot.

It became apparent that both these preliminary experiments had serious drawbacks. Firstly, only about 70% of the seed introduced germinated, which led to difficulties in any analysis of the results; secondly, more replication was necessary for them to be meaningful and thirdly, many millipedes in the pots did not feed on the seedlings since they were found to move deep into the soil.

Another experiment overcame some of these difficulties by ensuring close contact between millipede and seed, prolonging the period when seed are particularly vulnerable to attack, using a larger millipede to seed ratio and more replication and by using pre-germinated seed - all offered to the millipedes at a uniform stage of growth. Petri dishes permitted observations on the extent of aggregation around individual seeds and enabled dead individuals to be located and replaced.

Five pre-germinated seeds (cultivar 'Amono'), selected when the radicles were approximately 0.1 in. long, were pressed into the surface of a layer of compacted moist soil in each of five replicated Petri dishes for each of the following seed treatments: dieldrin,0.2%; methiocarb and mecarphon, 0.2% and 0.8% and PP505, 0.8%. Fifteen <u>B.guttulatus</u> were introduced into each dish. The soil was carefully compacted around the periphery of the arena so that the millipedes would be prevented from penetrating into the soil at the soil/glass face. The soil, previously oven-dried to kill any existing pests, was moistened to approximately 15-20% moisture content. Evaporation of water vapour was largely prevented by fitting a moist filter paper pad into the lid of each Petri dish and storing the Petri dishes in plastic boxes where a high humidity was maintained. The number of live millipedes around each seed was counted at 24 hr intervals and any dead millipedes recorded and replaced. After five days at 6-12°C the seedlings were removed, scored for damage on a 0-5 scale and replaced by fresh pre-germinated seed. A total of six successive sowings for each treatment were observed. observations on millipede mortality were made over a total period of 74 days until a treatment was found to have killed as many millipedes as was originally introduced (the 100% mortality level).

9.3. RESULTS

The data relating to both pest numbers, seedling damage and seedling establishment was analysed by Analysis of Variance; asterisks were used to denote whether any treatment mean differed significantly from the control.

9.3.1. Millipedes

The numbers of live and dead millipedes in the root zone of treated seed did not differ significantly from that of the control at either Welney or Boston sites (table 40). At Shouldham, although one eighth as many live B.tenuis were recovered from plots treated with gamma-BHC spray compared with the control, the result was not significant. The number of millipedes per core varied from 0 to 126 from the control plots and from 0 to 27 per core from plots treated with the gamma-BHC.

9.3.2. Pygmy beetle

Samples taken in 1971 indicated that seedlings grown from either treated seed or in insecticide-treated rows usually had more beetles in their root zones than in the root zones of seedlings from control plots, (table 41). The significantly higher numbers of both 'whole' and 'sections' of pygmy beetle recovered from the 1971 Boston samples on the gamma-BHC treated plots showed that a high proportion of the beetles had died in the root zone prior to sampling. In 1972

TABLE 40 SEED AND OVERALL SPRAY TREATMENT EFFECTS ON NUMBERS OF MILLIPEDES AROUND SEEDLINGS

Number of millipedes per seedling root zone

Treatment	Site / Insecticide	Welney 1971 (Live and dead)	Boston 1971 (Live and dead)	Shouldham 1972 (Live)
	control	2.6	1.1	32.1
Seed	dieldrin, 0.2%	0.1	-	-
	methiocarb, 0.8%	0.1	3.1	-
	gamma-BHC, 0.2%	0.5	-	-
Overall spray	gamma-BHC, 16oz. ai./ac.	0.6	0.8	4.4
Standard error of the difference between means (S.E.D.) ±		1.6	1.9	18.7
	Degrees of freedom	12	6	3

TABLE 41 SEED, FURROW AND OVERALL SPRAY TREATMENT EFFECTS ON NUMBERS OF PYGMY BEETLES AROUND SEEDLINGS

Number of beetles per seedling root zone

Treatment	Site / Insecticide	Boston 1971 (Live + dead)	Boston 1971 (Live + dead) (W)†	Boston 1971 (Live + dead) (S)†	Welney 1971 (Live + dead)
	Control	5.0	3.0	0.1	5.3
Seed	dieldrin, 0.2%	-	-	-	9.1
	methiocarb, 0.8%	3.3	-	-	9.4
	mecarphon, 0.8%	-	-	-	-
	PP505, 0.8%	-	-	-	-
	gamma-BHC, 0.2%	-	-	-	11.6
Furrow	methiocarb, 10oz. ai./ac.	-	8.1	1.4	-
	gamma-BHC, 4oz. "	-	-	-	-
	" 10oz. "	-	11.1*	2.8*	-
	" 16oz. "	-	-	-	-
	oxamyl, 10oz. "	-	-	-	-
	carbofuran, 10oz. "	-	-	-	-
Overall spray	gamma-BHC, 16oz. "	3.4	-	-	6.3
	S.E.D. $\pm$	1.8	2.7	0.8	3.6
	Degrees of freedom	6	6	6	12

† Whole, W and sections, S of beetles

*, **, *** denotes treatment means significantly different from control at P < 5%, 1%, 0.1% respectively.

when living and dead beetles were estimated separately from the Benwick sample, up to 91% of the beetles in the root zone were dead compared with only 2.6% from the control. Significantly more of both live and dead beetles were recovered from gamma-BHC treated rows and significantly more dead beetles from carbofuran treated rows compared with the control. Oxamyl was markedly less toxic to the pygmy beetles than the other insecticides. At Littleport and Terrington, numbers of both live and dead beetles around seedlings from both treated and untreated seed were comparatively few and although many more dead beetles were recovered from rows treated with gamma-BHC the difference was not significant.

9.3.3. Collembola

In 1971 few *Onychiurdae* were found around both treated and untreated seedlings and differences were not significant (table 42); in 1972 significantly more were found around seedling roots in rows treated with gamma-BHC but live and dead were not separated. At Shouldham, the gamma-BHC spray significantly reduced numbers of live *Onychiurus* in the root zone, whilst at Littleport in 1973 and at Stalham in 1974, gamma-BHC, either as a furrow or spray treatment, increased significantly the number of dead *Onychiurus* around seedling roots. Although the two seed treatments, tested at Stalham, decreased numbers of live and increased slightly the numbers of dead individuals in rows with treated seed, differences were not significant.

TABLE 42 SEED, FURROW AND OVERALL SPRAY TREATMENT EFFECTS ON NUMBERS OF COLLEMBOLA AROUND SEEDLINGS

Number of onychiurid Collembola per sample unit

Treatment	Insecticide (Site)	Boston 1971 (Live + dead)	Boston 1971 (Live + dead)	Welney 1971 (Live + dead)	Benwick 1972 (Live + dead)
	control	1.9	4.0	3.1	4.2
Seed	dieldrin 0.2%	-	-	-	-
	methiocarb 0.8%	3.7	-	2.9	-
	mercaphon 0.8%	-	-	-	-
	PP505 0.8%	-	-	-	-
	gamma-BHC 0.2%	-	-	1.5	-
Furrow	methiocarb, 10oz. ai./ac.	-	2.1	-	-
	gamma-BHC, 10oz. "	-	2.9	-	13.8***
	" 16oz. "	-	-	-	-
	oxamyl, 10oz. "	-	-	-	5.4
	Carbofuran, 10oz. "	-	-	-	7.2
Overall spray	gamma-BHC, 16oz. "	1.1	-	1.8	-
S.E.D. ±		1.3	1.5	1.3	2.3
Degrees of freedom		6	6	12	6

*** denotes treatment means significantly different from control at $P < 0.1\%$

continued on P. 286

TABLE 42 (CONTINUED)

Treatment	Site / Insecticide		Shouldham 1972 (Live)	Littleport 1973 (Live)	Littleport 1973 (Dead)	Stalham (Live)	Stalham (Dead)	Stalham . (Live)	Stalham . (Dead)
	control		11.7	11.1	0	41.0	21.4	17.2	13.4
Seed	dieldrin	0.2%	-	-	-	30.4	26.2	-	-
	methiocarb	0.8%	-	8.0	0	35.4	30.2	-	-
	mecarphon	0.8%	-	3.1	0	-	-	-	-
	PP505	0.8%	-	-	-	-	-	-	-
	gamma-BHC	0.2%	-	-	-	-	-	-	-
Furrow	methiocarb 10oz. ai./ac.		-	-	-	-	-	-	-
	gamma-BHC 10oz. ai./ac.		-	-	-	-	-	-	-
	gamma-BHC 16oz. ai./ac.		-	4.3	15.5*	-	-	-	-
	oxamyl 10oz. ai./ac.		-	-	-	-	-	-	-
	carbofuran 10oz. ai./ac.		-	-	-	-	-	-	-
Overall Spray	gamma-BHC 16oz. ai./ac.		6.4*	-	-	-	-	30.0	67.1***
S.E.D. ±			1.3	4.8	3.8	8.8	6.0	9.1	2.4
Degrees of freedom			3	9	9	33	33	3	3

*, *** denotes treatment neams significantly different from control at $P < 5\%$, 0.1% respectively.

9.3.4. Symphyla and wireworms

These two pests were relatively uncommon, occurred at only two sites and were present in small numbers. At Littleport both seed treatments and the gamma-BHC furrow treatment reduced numbers of live Scutigerella immaculata around the seedling roots but differences were not significant; numbers of dead symphylids around the seedling were only slightly different from the control (table 43). At Cottenham, wireworms extracted were not separated into live and dead but more were found in treated than untreated rows; differences between means were non-significant.

9.3.5. Mites

The soil-inhabiting mites recovered during 1971 and 1972 were identified as: Rhodocarus roseus (Oudemans), Parasitus lunaris (Berlese), Macrocheles sp., Hypoaspis acefier (Canestrini), Veigaia planicola (Berlese), Rhagidia sp. and Tyrophagus sp. In 1974 the principal mite recovered from the Stalham site was Pergamasus sp. Gamma-BHC was the only material which affected numbers in the root zone of the seedlings, significantly increasing numbers of dead individuals in treated rows at Stalham (table 44). Gamma-BHC also decreased the number of live mites in the seedling root zone at Stalham and at Shouldham and significantly decreased numbers around seedling roots at Boston when live and dead were not separated. The methiocarb furrow treatment also decreased numbers in the root zone.

TABLE 43 SEED AND FURROW TREATMENT EFFECTS ON NUMBERS OF SYMPHYLA AND WIREWORMS AROUND SEEDLINGS

Number of Symphyla (Littleport) and wireworms (Cottenham) per sample unit

Treatment	Site / Insecticide	Littleport 1973 (Live)	Littleport 1973 (Dead)	Cottenham 1974 (Live + dead)
	control	6.9	1.5	0.8
Seed	methiocarb, 0.8%	2.6	1.9	-
	mecarphon, 0.8%	1.4	0.5	-
Furrow	gamma-BHC, 4oz. ai./ac.	0.5	2.0	-
	oxamyl, 4oz. "	-	-	1.2
	" 16oz. "	-	-	1.6
	aldicarb, 4oz. "	-	-	1.8
	" 16oz. "	-	-	2.0
	S.E.D. $\pm$	2.9	0.6	1.0
Degrees of freedom		9	9	12

TABLE 44 FURROW AND OVERALL SPRAY TREATMENT EFFECTS ON NUMBERS OF MITES AROUND SEEDLINGS

Number of mites per sample unit

Treatment	Site / Insecticide	Boston 1971 (Live + dead)	Benwick 1972 (Live + dead)	Shouldham 1972 Live	Stalham 1974 Live	Stalham 1974 Dead
	control	2.3	4.3	12.6	1.2	1.7
Furrow	methiocarb, 10oz. ai./ac.	1.0*	-	-	-	-
	gamma-BHC, 10oz. "	1.0*	5.7	-	-	-
	oxamyl, 10oz. "	-	5.1	-	-	-
	carbofuran, 10oz. "	-	5.9	-	-	-
Overall spray	gamma-BHC, 16oz. "	-	-	6.3	0.8	4.7*
S.E.D. ±		0.4	2.2	2.4	0.6	0.6
Degrees of freedom		6	6	3	3	3

* denotes treatment means significantly different from control at $P < 5\%$

9.3.6. Seedling damage

The amount of damage to seedling roots was decreased by all insecticide treatments but only to a significant extent by oxamyl and gamma-BHC furrow treatments (Gedney, 1974) and by dieldrin and methiocarb seed-treatments at Stalham, 1974, (table 45). At Cottenham, the amount of damage to seedling roots by wireworms was scarcely different on treated or untreated rows.

9.3.7. Seedling establishment

Where a pest attack was particularly severe, as at Boston, Welney, Benwick and Terrington, most insecticide treatments significantly improved seedling establishment (table 46). At these sites pygmy beetle attack had resulted in percentage establishments of only 27, 38, 9, 0 and 3% respectively. At Cottenham, only aldicarb granules, at 16 oz. ai./ac., significantly improved establishment and the crop was later ploughed-in and resown. At Stalham, only dieldrin and methiocarb seed treatments significantly increased seedling establishment.

9.3.8. Laboratory tests with millipedes

In the first preliminary experiment all the seed treatments decreased the number of seedlings that were damaged (table 47). The damage in this experiment was limited to excision of radicle tips; the high rate of millipede mortality showed that the millipedes readily came into contact with the treated seed. In the second experiment

TABLE 45 SEED, FURROW AND OVERALL SPRAY TREATMENT EFFECTS ON DAMAGE TO SEEDLING ROOTS BY SOIL-INHABITING PESTS

Root damage assessment - mean damage score (0-5) per seedling

Treatment	Site / Insecticide	Cottenham 1974 wireworms	Gedney 1974 millipedes	Stalham 1974 Collembola	Stalham 1974 Collembola
	control	2.1	1.8	2.5	1.6
Seed	dieldrin, 0.2%	-	-	1.8*	-
	methiocarb, 0.8%	-	-	1.4***	-
Furrow	oxamyl, 4oz. ai./ac.	1.6	-	-	-
	" 8oz. "	-	1.5*	-	-
	" 16oz. "	1.6	-	-	-
	aldicarb, 4oz. "	2.0	-	-	-
	" 16oz. "	1.2	-	-	-
Overall spray	gamma-BHC, 16oz. "	-	1.1***	1.2	1.2
	S.E.D. ±	0.6	0.1	0.3	1.0
	Degrees of freedom	12	6	32	15

*, *** denotes treatment means significantly different from control at $P < 5\%$ and 0.1% respectively

TABLE 46 SEED, FURROW AND OVERALL SPRAY EFFECTS ON SEEDLING ESTABLISHMENT AT PEST-INFESTED SITES

Percentage seedling establishment.

Treatment	Insecticide (Site)			Boston 1971	Boston 1971	Welney 1971	Benwick 1972	Shouldham 1972
	control			27.2	38.2	8.8	0	60.9
Seed	dieldrin,	0.2%		-	-	8.3	-	-
	methiocarb,	0.8%		56.9***	-	47.0***	-	-
	mecarphon,	0.8%		-	-	-	-	-
	PP505,	0.8%		-	-	-	-	-
	gamma-BHC,	0.2%		-	-	31.2***	-	-
Furrow	methiocarb,	10oz.	ai./ac.	-	41.2	-	-	-
	gamma-BHC,	4oz.	"	-	-	-	-	-
	"	10oz.	"	-	-	-	-	-
	"	16oz.	"	-	-	-	23.6***	-
	oxamyl,	4oz.	"	-	-	-	-	-
	"	10oz.	"	-	-	-	11.9**	-
	"	16oz.	"	-	-	-	-	-
	carbofuran,	10oz.	"	-	-	-	43.6***	-
	aldicarb,	4oz.	"	-	-	-	-	-
	"	16oz.	"	-	-	-	-	-
Overall spray	gamma-BHC,	16oz.	"	65.6***	-	45.6***	-	69.1
S.E.D. ±				3.3	4.0	4.3	3.4	7.3
Degrees of freedom				45	99	45	81	51

continued on P. 293

*, **, *** denotes treatment means significantly different from control at P < 5%, 1%, 0.1% respectively

TABLE 46(CONTINUED)

Percentage seedling establishment

Treatment	Site / Insecticide	Little-port 1973	Terring-ton 1973	Cotten-ham 1974	Stalham 1974	Stalham 1974
	control	66.9	3.2	16.5	60.5	57.5
Seed	dieldrin, 0.2%	57.7	4.5	-	64.8*	-
	methiocarb, 0.8%	59.4	18.8***	-	68.2***	-
	mecarphon, 0.8%	62.6	11.8**	-	-	-
	PP505, 0.8%	-	16.9***	-	-	-
	gamma-BHC, 0.2%	-	-	-	-	-
Furrow	methiocarb, 10oz. ai./ac.	-	-	-	-	-
	gamma-BHC, 4oz. "	-	3.7	-	-	-
	" 10oz. "	-	-	-	-	-
	" 16oz. "	75.0	26.0***	-	-	-
	oxamyl, 4oz. "	-	-	22.0	-	-
	" 10oz. "	-	-	-	-	-
	" 16oz. "	-	-	26.7	-	-
	carbofuran, 10oz. "	-	-	-	-	-
	aldicarb, 4oz. "	-	-	28.0	-	-
	" 16oz. "	-	-	28.7*	-	-
Overall spray	gamma-BHC, 16oz. "	-	-	-	-	64.3
	S.E.D. ±	7.2	2.6	5.8	2.1	4.5
	Degrees of freedom	43	48	16	68	32

*, **, *** denotes treatment means significantly different from control at $P < 5\%$. 1%, 0.1% respectively

TABLE 47 EFFECT OF PELLETED SEED TREATMENTS ON MILLIPEDES AND SEEDLINGS IN TWO PRELIMINARY EXPERIMENTS IN THE LABORATORY

Observation		First experiment		Second experiment	
Insecticide		Proportion of seedlings with severe damage	% mortality of *B. superus* after 15.5 days	Damage score per seedling (0-5)	% mortality of *B. guttulatus* after 28 days
control		3/9	5	1.4	11
dieldrin,	0.2%	0/8	5	1.7	9
"	0.8%	0/8	15	-	-
methiocarb,	0.2%	1/9	15	1.8	0
"	0.4%	-	-	1.0	16
"	0.8%	0/9	50	2.0	20
mecarphon,	0.2%	-	-	1.2	27
"	0.4%	-	-	1.3	13
"	0.8%	-	-	1.1	15
PP505,	0.8%	-	-	1.3	31
carbaryl,	0.8%	0/9	85	-	-

seedling damage was assessed by a scoring method but the results were very inconsistent and all treated and untreated seed showed varying amounts of slight root damage (table 47). At the end of the experiment many millipedes were found at the bottom of the pots in close contact with the moist filter paper pad lining the base. When scored for damage the seedling roots were often up to 3 in. long and the feeding holes were usually either above or below the seed pellet zone.

The improved experiment used a total of 875 seedlings, each of which were scored for root damage. All seedlings from treated seed showed less root damage than the seedlings from the control and there was a tendency for higher rates to give slightly better protection than the low rates of each insecticide (table 48). The numbers of millipedes seen around the seed was approximately proportional to the amount of damage. Seed treated with mecarphon caused the highest millipede mortality and the control seed the least. Millipedes in contact with insecticide-treated pellets often displayed a red body colouration, resulting from the discharge from paired defence glands situated on the body segments; this usually accompanied an un-coordinated movement of the legs.

TABLE 48 EFFECT OF PELLETED SEED TREATMENTS ON MILLIPEDES AND SEEDLINGS IN THE LABORATORY

Observation Insecticide		Millipedes per seedling per 24 hr period	Mean damage score per seedling after 5 days (0-5)	% mortality of B.guttulatus after 74 days
control		0.95	1.26	3
dieldrin	0.2%	0.22***	0.70***	31
methiocarb	0.2%	0.28***	0.47***	23
"	0.8%	0.29***	0.37***	39
mecarphon	0.2%	0.19***	0.14***	47
"	0.8%	0.21***	0.11***	100
PP505	0.8%	0.13***	0.07***	44
S.E.D. ±		0.09	0.13	-
Degrees of freedom		30	24	-

*** denotes treatment means significantly different from control at $P < 0.1\%$

9.4. DISCUSSION

Millipedes are the most unpredictable soil-inhabiting pests and the ones that form the most closely compacted aggregates around seedling roots. The small infestations where experiments were sited has produced inconclusive information in the present study. At the Shouldham site the considerable variation in numbers between individual seedling roots, both on plots treated with gamma-BHC and untreated plots, produced a Standard Error sufficiently large to render the difference between means non-significant; a logarithmic transformation may have helped to stabilize the variance of this data. Other pests do not form such large aggregates around seedling roots and transformation of the data was unnecessary. The small (8%) and non-significant increase in seedling establishment on treated plots at Shouldham suggested that, although feeding aggregates were large, the millipedes were not causing extensive damage. That blaniulid millipedes may remain in contact with the root but not cause damage has been observed in feeding studies in the laboratory and reported in a previous section. Insecticides, in a seed treatment formulation, have usually given only small benefits in protecting seedlings against millipedes when tested in field experiments. At Bottisham in 1971, where B.tenuis was causing damage, all seed-treatments decreased seedling numbers slightly but in addition field-mice were causing seed loss (Dunning and Winder, 1972). Experiments in 1973, at two sites having millipede infestations, Kettering and Marham, showed that the most effective seed treatment (methiocarb, 0.2%) could increase seedling establishment by 15% and 19% respectively at the two sites. The standard dieldrin treatment increased establishment by only 5% and 4% respectively (Dunning and Winder, 1974). In the laboratory the pot experiment showed that millipedes may cause as much damage to seedlings from treated as from untreated seed if the root system is sufficiently developed to allow them to feed on areas some distance from the pellet zone; the results of this experiment were inconclusive owing to the difficulties of ensuring that the millipedes would remain in the vicinity of the root. Experiments using millipedes in Petri dishes showed that some materials can give good protection under those conditions which increased the ease of access of the pellets to the millipedes and ensured more contact. As in field experiments, dieldrin (0.2%) was the least effective insecticide and mecarphon (0.8%) and PP505 (0.8%) the most effective. At Kettering, however, these two materials were amongst the least effective but at Marham they were amongst the best. The difference in their performance at these two sites cannot readily be explained. Gamma-BHC seedbed spray, at 16 oz. ai./ac., increased seedling establishment by 36% and 10% at Kettering and Marham respectively (Winder and Dunning, 1974); this indicates that, at the first site, the seed treatments were giving relatively little protection, whilst at Marham the best seed treatments were more effective than the gamma-BHC. Generally, the evidence is conflicting as to whether seed-treatment is as effective as an overall spray of gamma-BHC at millipede-infested sites but the choice may depend on the size of the infestation in the seedbed. In adjacent seed spacing experiments at these two sites more millipedes were recorded in the root zone of untreated seedlings at Marham at three sampling times from April until June. The relatively good performance of seed treatments at Marham would lead one to have expected the opposite but other factors such as the numbers of Onychiurus in the root zone may be involved, in addition to the size of the millipede infestation.

Other compounds also performed inconsistently. In a previous experiment and reported in another section oxamyl granules (16 oz. ai./ac.) significantly increased seedling numbers on plots left uncompacted but in the present study oxamyl at 8 oz. ai./ac. did not have a significant benefit but seedling establishment at the site was generally poor due to unusually dry seedbed conditions.

D.D.T. was not tested in the present study because of its long persistence in the soil but this material has been found to be particularly effective against millipedes (Dempster, 1967; Edwards, 1974).

Most seed treatments tested, but not dieldrin, and both furrow and overall sprays, especially gamma-BHC, protect seedlings adequately against pygmy beetle. Soil sampling in 1971 suggested that the increased numbers of beetles in the root zones on effective treatments probably includedmany dead beetles. Further sampling, in 1972, where live and dead beetles were separated, confirmed this suggestion. Soil sampling showed that furrow-treatments, particularly gamma-BHC, are the most effective method of killing pygmy beetles.

Collembola, particularly Onychiurus armatus, were found at most sites together with the other pests but until recently their ability to cause damage independently has been shown to be underestimated. Numbers in the soil at the different sites varied considerably; there were few at Welney and Boston but many at Littleport, Shouldham, Kettering and Stalham, though at Kettering they were comparatively few compared with the number of millipedes. Gamma-BHC, either as a furrow or overall spray treatment, was the only material which proved to be significantly active against

Onychiurus. Although both dieldrin and methiocarb significantly decreased damage and increased seedling establishment, differences were not reflected in the numbers of *Onychiurus* in the root zone of the seedlings; it is possible that these insecticides were exerting a repelling effect. Heijbroek (1972) noted that seed furrow treatment with lindane (gamma-BHC) resulted in large concentrations of *O.armatus* in the beet rows and ascribed this to the attractiveness of the insecticide to the *Onychiurus*. These results may be due partly to a sub-lethal effect of the insecticide on *Onychiurus* which may serve only to restrict locomotion. Many *O.armatus* from gamma-BHC treated plots in the present study showed such symptoms and were classified as 'dead' if such un-coordinated movement prohibited locomotion; those from untreated plots were more readily definable as either 'live' or 'dead'. That many of these 'un-coordinated' *Onychiurus* were probably incapable of normal feeding is implied in the small non-significant increase in seedling numbers on gamma-BHC treated plots.

In the laboratory, mites, (*Pergamasus* sp.), have been shown to feed readily on *O.armatus*. These mites were sensitive to gamma-BHC; this insecticide may have a long term effect on *Onychiurus* numbers in the soil but in the present study this aspect was not investigated. At two sites in 1974 numbers of the prostigmatid mite, *Allothrombium fuliginosum* on the foliage of sugar-beet plants were significantly fewer on plots receiving aldicarb in the seed furrow than on untreated plots (Baker and Dunning, 1975). The studies provided no conclusive evidence that Symphyla or wireworms were particularly sensitive to the insecticides tested since numbers in the soil were relatively few. At Littleport, where *Scutigerella immaculata* was the main pest, although *Onychiurus* spp. was also present, none of the treatments significantly affected seedling establishment. At Cottenham, where the attack was due almost entirely to wireworms, the insecticides scarcely affected the amount of damage to the seedling roots nor was seedling establishment increased on all insecticide-treated rows; this may be due to the wireworms having fed on the roots before succumbing to the insecticide.

All the results emphasise that dieldrin, applied to all commercial seed until 1973, is ineffective against most soil-inhabiting pests. Its possible replacement may be methiocarb which has given good protection against pygmy beetle attack and sometimes where millipedes have been present. It is still difficult to determine to what pest most damage can be attributed if a mixture is present in the soil; this task is now made even more difficult since it has been shown that Onychiurus spp. itself is able to cause seedling loss. Morris (1927), in his census of different arthropod groups in arable soil, has found that Collembola are the most numerous group, of which O.armatus was sometimes the most common arthropod species. Perhaps, if more attention was given to the protection of seedlings from this group of pests, seedling establishment may be increased in areas where pest problems were not previously recognized.

Gamma-BHC overall spray or furrow treatment still remains the standard by which to compare seed treatments but a better understanding of its effects on predators of soil-inhabiting pests is necessary. Also the effects of any replacement non-organochlorine compound on predators must be considered and long term effects on pest numbers monitored.

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APPENDIX TABLE 1. SEEDLING SPACING AND PEST POPULATIONS IN THE ROW AND SOURCES OF DATA FOR EACH PEST FROM WHICH MEAN VALUES WERE CALCULATED.

	A Millipedes			B Pygmy beetle			C Collembola		
Seedling spacing (in.)	1.5	4.5	9.0	1.5	4.5	9.0	1.5	4.5	9.0
Number of pests per seedling (mean from four sites - see below)	2.3	5.0	6.0	0.9	1.2	1.9	2.5	2.7	2.7
Seedlings/acre (thou.)	209	70	35	209	70	35	209	70	35
Estimated pests/acre in the root zone (thou.)	481	350	210	188	84	67	523	189	95

A Data from Shouldham 6.6.72
Marham 17.5.73
Kettering 14.5.73
Bottisham 14.5.73

B Data from Shouldham 6.6.72
Kettering 30.5.73
Littleport 2.5.73, 24.5.73
Marham 17.5.73, 6.6.73

C Data from Broom's Barn 17.4.73
Kettering 25.4.73
Littleport 2.5.73
Stalham 6.5.74

APPENDIX TABLE 2. THE PRESENCE OF MILLIPEDES ON DIFFERENT SOIL TYPES

Soil type	Fields searched			Fields with millipedes		
	1970	1971	% mean both years	1970	1971	% mean both years
Loamy coarse sand	18	8	4.5	3	0	3.5
Loamy sand	6	8	2.4	0	1	1.2
Loamy v.f. sand	7	16	4.0	0	0	0
Coarse sandy loam	23	19	7.3	2	0	2.4
Sandy loam	78	73	26.1	12	8	23.5
v.f. sandy loam	19	22	7.1	2	5	8.2
Loam	23	32	9.5	5	6	12.9
Silty loam	27	31	10.0	7	2	10.6
Sandy clay loam	37	32	11.9	4	4	9.4
Clay loam	17	29	8.0	9	4	15.3
Clay	8	9	2.9	1	2	3.5
Light peat	6	7	2.2	2	0	2.4
Loamy peat	5	11	2.8	1	2	3.5
Peaty loam	1	5	1.0	1	1	2.4
Others	1	0	0.2	1	0	1.2

APPENDIX TABLE 3. AGGREGATION AND DAMAGE TO RAW AND PELLETED SEED BY BLANIULUS AND ONYCHIURUS IN THE LABORATORY.

Number of pests per 10 seeds (P = pelleted; R = raw)

			Sowing 1	2	3	4	5	6	7	8	$\bar{x}$ pests /sowing	L.S.D. P = 5%		P
a	Blaniulus	P	11.0	8.0	9.7	4.3	14.0	8.8	18.2	8.3	10.2	3.7	a-b	n.s.
b		R	11.3	12.0	5.2	3.8	8.5	8.5	5.3	10.2	8.4		a-c	*
c	Onychiurus	P	4.8	7.2	7.8	8.7	5.0	1.7	7.2	6.0			a-d	n.s.
d		R	7.0	11.5	9.2	3.8	4.0	7.5	2.8	11.2	6.9		b-c	n.s.
													b-d	n.s.
													c-d	n.s.

$\bar{x}$ pests / seed / Sowing

a All seed		b All pests	
Blaniulus	9.3	Raw seed	7.6
Onychiurus	6.4	Pelleted seed	8.1
L.S.D. 5%*	2.6	L.S.D. 5%*	2.6
P	*	P	n.s.

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APPENDIX TABLE 3. (CONTINUED)

Mean damage score (0 - 5) per seedling

			Sowing 1	2	3	4	5	6	7	8	$\bar{x}$ pests /sowing	L.S.D. P = 5%		P
a	*Blaniulus*	P	2.7	0.9	0.7	0.4	0.4	1.2	1.2	0	1.0	0.8	a-b	n.s.
b		R	1.6	1.0	0.3	0.5	0	1.4	0.1	0.3	0.7		a-c	n.s.
													a-d	n.s.
c	*Onychiurus*	P	1.4	1.7	2.4	2.0	0.4	1.9	1.2	2.2	1.7		b-c	n.s.
d		R	1.6	3.1	1.6	2.7	1.9	3.7	1.1	3.6	2.4		b-d	***
													c-d	n.s.

$\bar{x}$ damage score / seed / Sowing

a All seed		b All pests	
Blaniulus	0.8	Raw seed	1.5
Onychiurus	2.0	Pelleted seed	1.3
L.S.D. 5%*	0.5	L.S.D. 5%*	0.5
P	*** (P <0.1%)	P	n.s.

PUBLICATIONS

The following papers are included in this thesis for consideration. Those concerned with soil-inhabiting pests represent condensed versions of the work reported earlier in this thesis; the study of the Carabidae was mainly subsidiary to that on pests :

BAKER, A.N. (1974).
Some aspects of the economic importance of millipedes.
Symp. zool. Soc. Lond. **32**, 621-628

BAKER, A.N. (1975).
Association of populations of onychiurid Collembola with damage to sugar-beet seedlings.
Pl. path. (In Press).

BAKER, A.N. & DUNNING, R.A. (1975).
Some effects of soil type and crop density on the activity and abundance of the epigeic fauna, particularly Carabidae, in sugar-beet fields.
J. app. Ecol. (In Press).

DUNNING, R.A., BAKER, A.N. & WINDLEY, R.F. (1975).
Carabids in sugar-beet fields and their possible role as aphid predators.
Ann. Appl. Biol. **80**, 125-128

Symp. zool. Soc. Lond. (1974) No. 32, 621–628.

SOME ASPECTS OF THE ECONOMIC IMPORTANCE OF MILLIPEDES

A. N. BAKER

Broom's Barn Experimental Station, Higham, Bury St. Edmunds, Suffolk, England

SYNOPSIS

Millipedes, especially *Brachydesmus superus* (Latz.) and *Blaniulus guttulatus* (Bosc.) can stunt and even kill sugar-beet seedlings in the spring by their aggregated feeding on the young roots. Fortunately, seedlings are at risk over only a limited period, up to about mid-June, after which the millipedes retreat deeper into the soil. The intensity of damage from millipedes varies considerably from year to year and is difficult to forecast, being largely dependent both on rate and vigour of seedling growth and on the numbers and activity of the millipedes. Present investigations, both in the laboratory and in the field, aim at a better understanding of the movements of populations in the soil in order to develop means of protecting the germinating seedling until it is established.

INTRODUCTION

The undesirable effect of millipedes on cultivated plants has been recognized for at least 80 years, but the true nature of the relationship is still controversial; they feed not only upon decaying organic material but also, under some circumstances, upon living plant tissue; Ormerod (1890) observed *Blaniulus guttulatus* attacking strawberry fruits and germinating mangold seed in 1885. Millipede feeding habits were investigated by Brade-Birks (1930), and Rolfe (1937–39) listed species and reported millipede damage to a wide range of horticultural and field crops including beans, peas, cucumbers, cabbage, cereals, lettuce, potatoes and sugar-beet; Wilson (1943) commented that, although they are usually secondary pests, millipedes will feed on the skin of soft skinned varieties of potatoes. Baurant (1964) has described the damage to sugar-beet seedlings attributable to two blaniulid species in Belgium. Spring-sown crops are particularly prone to attack because they are at their tenderest stage when soil conditions are conducive to millipede activity. Warm, moist, glasshouse soils ensure large populations and intense activity throughout the year and *Oxidus gracilis* (Koch), a tropical species, is common in glasshouses both in the British Isles and in south and west U.S.A. (Edwards & Gunn, 1961; Henneberry & Taylor, 1961).

This paper deals primarily with the damage and field behaviour of the most common species that occur where sugar-beet is grown in eastern England and is part of the results from a three-year study on some soil-inhabiting pests of sugar-beet.

MILLIPEDES AS PESTS OF SUGAR-BEET SEEDLINGS

Millipedes cause loss of sugar-beet seedlings; this is especially important with current agricultural trends to minimize labour by sowing fewer seeds at wide spacing and by increased herbicide usage which eliminates any other competing seedlings of other species. Monogerm sugar-beet seed is sown in March or April, between three and eight in. apart in rows 20 in. apart. At this time temperatures in the seedbed may range from 5°C to 7°C and up to five weeks may elapse before half the seeds have germinated. If millipedes are active in the seedbed they may feed on the emerging radicle, unfolding cotyledons and hypocotyl below soil level, making lesions which sometimes coalesce and cause complete separation of these parts, often resulting in death of the seedling. More usually, however, millipedes attack later when the cotyledons are above ground; root damage impairs the seedling's ability to extract water and nutrients from the soil and, if severe, may kill the plant. Nutrient deficiency and wilting are common symptoms of millipede damage to roots (Baker, 1971a).

THE IMPORTANT SPECIES

Advisory Entomologists report that blaniulids cause most damage, especially *Blaniulus guttulatus* (see Cloudsley-Thompson, 1950) and sometimes *Boreoiulus tenuis* (Bigler) and *Archeboreoiulus pallidus* (Brade-Birks). The polydesmids are also sometimes implicated, especially *Brachydesmus superus* which is very common in arable fields and gardens.

A consistent and important feature of the behaviour of millipedes, and of other soil-inhabiting pests of sugar-beet seedlings, is their tendency to move into the seed rows after sowing and to aggregate around the seedlings. This is particularly noticeable with the blaniulids and up to 121 per seedling have been recorded. Soft plant materials are the best food source for millipedes and seedlings are susceptible to damage for a limited period; this usually extends from germination to the four-leaf stage—a period of eight weeks at the most.

Damage is usually most severe in May and is rare by July. Soil conditions conducive to rapid germination and growth also encourage movement and feeding of millipedes in the seedbed. During July the millipedes move deeper into the soil and any fresh damage to the tap-root is unlikely. *B. superus and B. guttulatus* differ not only morphologically but also in their field behaviour, and their relative importance as pests reflects these behavioural differences.

BRACHYDESMUS SUPERUS: A SOIL SURFACE SPECIES

B. superus, the commonest polydesmid of arable land, is a most active animal and lives in the loose topsoil except when moulting. If undisturbed it can be found throughout the year near the soil surface or under surface vegetation, but ploughing the land early in winter buries most of the millipedes and they are then difficult to find. In late winter this millipede sometimes aggregates around ploughed-in cereal stubble, manure, or sugar-beet crowns which have decayed enough to be attractive. *B. superus* may cause damage to early-sown sugar-beet before the arrival of the blaniulids in the seed bed from the deeper soil.

VERTICAL DISPLACEMENTS OF THE BLANIULIDAE

The depth distributions in sugar-beet fields of the blaniulids, *B. guttulatus* and *A. pallidus*, have been extensively studied in Belgium (Pierrard, Bonte & Baurant, 1963; Biernaux & Baurant, 1964). Successive 10 cm deep soil samples provided some evidence of a vertical displacement, apparently controlled by differences in soil temperature and moisture; Dowdy (1943–1944) and Blower (1970) also mention vertical movements of millipedes in response to temperature. Most of the blaniulids were found in the 30–90 cm deep soil zone in winter but by the end of April, the time when attacks on sugar-beet seedlings first occur, most are in the top 30 cm. This upward movement of blaniulids has been shown to start when the temperature of their surroundings rises above 4–6°C (Pierrard *et al.*, 1963). During the summer months they move downwards from the top soil; Biernaux (1968) demonstrated their avoidance of soil with a moisture content of less than 12%. After a brief reappearance near the soil surface in the autumn, when the soil is remoistened, they retreat to deeper soil layers.

AGE-STRUCTURES, ACTIVITY AND DAMAGE

B. superus is an annual; the adults produce one brood and then die (semelparity). The duration of moulting, the time spent in each stadium, and the longevity of adults are determined by the temperature of the habitat. Because they are surface-inhabiting animals, they will be directly influenced by seasonal temperature fluctuations. Populations have relatively narrow age-structures. In two consecutive years, populations in the autumn have had a preponderance of adults (64% in December at one site) with a sex ratio of up to three females to one male. Mating can be readily observed in September and October on the soil surface, in soil crevices and under leaves. *Brachydesmus* nests, with egg clusters, have been found occasionally on compacted soil at plough depth in January but oviposition occurs most readily in Spring. No hatching occurs at 2–4°C, but takes only about 20 days at 15–20°C. The threshold value for hatching, in the region of 7–11°C, agrees well with the occurrence of stadium I in soil samples taken in April and May when these temperatures exist in the top 10 cm of soil. At 15–20°C larvae attain stadium V within three months, whereas they fail to moult at 2–4°C. The adult and penultimate stadia are the most harmful to plants and there is evidence that severe damage occurs only when seedling germination coincides with a large adult population; thus factors which control the rate of development of *Brachydesmus* can affect its potential as a pest (Baker, 1972a).

B. guttulatus adults may breed in successive years (iteroparity), producing a wide spectrum of ages in the population. This millipede may eventually reach stadia XVI or XVII; assuming three moults per year the calculated life span could be five or six years (Biernaux, 1967b). Field observations suggest both spring and autumn oviposition peaks (Biernaux, 1967b; Kinkel, 1955). Information about the rate of development indicates that larvae from eggs laid at the end of April can reach stadium V by October and then overwinter; the adult stage is reached only in the second year of growth (Biernaux, 1967a). All stadia, except I, of *Blaniulus* have been observed feeding upon sugar-beet seedlings and they may be found in the seedbed from March until late June (Baker, 1971a; Biernaux, 1967a).

WORK IN PROGRESS ON MILLIPEDE BEHAVIOUR

Studies of behaviour in the laboratory may help in understanding what happens in the field. My investigations suggest that to interpret millipede movements solely as a response to variations in temperature and moisture may be an over-simplification of the relationship between millipedes, plants, and their common environment.

Laboratory studies support field evidence that an increase in soil temperature, similar to that in the field in spring, leads to increased activity, an increase in numbers of larvae and a general dispersal.

Some millipedes move upwards from the sub-soil into the seedbed; here, large diurnal temperature fluctuations ensure intense activity. In the laboratory activity is initiated by both increasing and decreasing temperature but constant temperatures have a depressing effect. The millipedes aggregate around the fruit coat as soon as the radicle and embryo has become exposed on germination. When excised parts of seedlings are offered separately they feed preferentially upon young cotyledon tissue; they also aggregate and feed upon 5% agar discs but prefer those incorporating 5% glucose or 5% sucrose solutions (Baker, 1971a).

Conditions which cause millipedes to stop feeding are important because the longer they feed, the greater the damage they cause. In warm soil they tend to moult more frequently, and therefore stop feeding and leave the surface root zone to build moulting chambers deeper in the soil. Also, increasing plant maturity is associated with a dispersal from the seedbed. Laboratory experiments demonstrate a sensitivity both to soil moisture and to low temperatures, the latter resulting in a positive geotaxis and aggregation in the deepest penetrable soil layers; similar effects can be observed in the field.

CONCLUSIONS

For millipedes to cause significant damage to seedlings they must be sufficiently numerous, sufficiently active to aggregate around the germinating seeds and they must find the seedlings an attractive food source on which to continue feeding.

It is difficult to decide what conditions lead to the build-up of large populations. Recent surveys of millipede populations during the autumn suggest that soil type and geographical location, especially in respect of topography, exert an influence. Millipedes, mainly *B. superus* but some *B. guttulatus*, were found most frequently on the heavier soils—clay loams, silts and heavy peats (Baker, 1971b, 1972b). They were particularly common in certain low lying regions of East Anglia near rivers and estuaries. Little is known of the biological regulation of millipede populations in the field, but there is some evidence that the predatory soil mite *Pergamasus quisquiliarum* Canestrini feeds upon stadium I *B. superus*.

Resistence of a seedling to attack depends upon both its genetic make-up and the conditions which govern the rate of growth. Millipede activity appears to be under the control of edaphic factors, mainly temperature and moisture; it is the most difficult aspect of the syndrome to control, since it is largely governed by climate. However, compacting the soil in order to limit the space for movement in the root zone seems a promising, though by no means new, approach to decrease activity.

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Discussion

Enghoff: Have you ever found *Boreoiulus tenuis* as a pest of sugar beet?

Baker: Yes, it has been identified as the main seedling pest at Shouldham, near Downham Market, Norfolk, and at Bottisham, Cambridgeshire, both shallow, chalky loam soils over chalk. It does exactly the same sort of damage as the other blaniulids—but in general it is less common.

Biernaux: In Belgium, we have *Archeboreoiulus pallidus* and *Boreoiulus tenuis* together with *Blaniulus guttulatus.*

Jeekel: I think it was Cloudsley-Thompson (1950)* who postulated the idea that a period of drought would increase the severity of millipede damage; he suggested the millipedes would attack the crop to find water. Does this conflict with your idea that they go down when it is dry?

Baker: I have shown in the laboratory that millipedes will damage seedlings even in soil with suitable moisture content, when they are not suffering from a water deficit. I find it hard to accept the hypothesis that they attack plants to find water. I think that millipedes become active as the result of increased soil temperatures, after the prolonged winter inactivity. They start moving; an individual comes across a seedling and attacks it. There seems to be some attraction between millipedes; once one has started to feed, others come along and they form a swarm; the sugars, too, may help to maintain their interest in the plant. Of course when a plant, especially a young seedling, is being deprived of water then growth is restricted, and because it can neither mature enough to harden its root tissue against millipede attack, nor compensate by growing replacement water- or nutrient-absorbing tissue, the seedling would therefore be expected to succumb more easily to a given intensity of millipede attack under "dry" soil conditions. My conclusions, as a result of field and laboratory observations, only go as far as proving that millipedes can cause primary damage to seedlings when the seedlings are grown in soil with adequate moisture for growth. I have not been able to compare directly the amount of damage

* See list of references, p. 626.

done to seedlings grown in "dry" versus "wet" soils. But I have done an experiment where 5% agar discs incorporating a sugar were substituted for seedlings and found no significant difference in the amount of material eaten by the millipedes. I therefore find it hard to accept the hypothesis put forward.

BRADE-BIRKS: Do you have any idea how an attack begins? Often a crop remains unattacked one year and then there is a big influx of millipedes the next. Is it because climatic conditions in spring are suitable for the development of eggs or because the introduction of manure and organic waste material from the factories provides the right conditions for development?

BAKER: I think both these factors may be important, but since 1935 the growing of sugar beet in successive years was prohibited following the introduction of a clause in factory contracts; there is usually a three or four year gap between the growing of beet during which time the land is under cereals.

TURNER: Do millipedes transmit viruses?

BAKER: I do not know. I would not have thought they would be ideal vectors because blaniulids, at least, are not very mobile.

HEATH: On what types of soil do you get the most severe attacks?

BAKER: A severe attack usually occurs on land which supports large numbers of millipedes and which usually has a history of millipede trouble—land which is low lying, has a high organic content and a high cereal stubble content. The soils usually tend to be the silty loams, silts and the heavier peats; the soils must be porous to allow the millipedes to escape adverse conditions; these are the features of a good millipede soil!

Association of Populations of Onychiurid Collembola with Damage to Sugar-beet Seedlings

by A.N. BAKER and R.A. DUNNING

Broom's Barn Experimental Station, Higham, Bury St.Edmunds, Suffolk

S U M M A R Y

In eastern England *Onychiurus armatus* (Tullb.) and *O. fimetarius* (L.) were found associated with damaged sugar-beet seedling roots soon after sowing in mid-March, 1973, when the mean soil temperature ranged from 3 to 11°C; observations confirmed that *O. armatus* caused similar damage to seedlings under laboratory conditions. In field experiments *Onychiurus* spp. were always more numerous in the seedling root zone than between the seedling rows, but different seed spacings had no significant effect on the extent of aggregation around individual sugar-beet seedlings.

I N T R O D U C T I O N

In Holland, Belgium and W. Germany *Onychiurus* spp., particularly *O. armatus* (Tullb.), are regarded as important pests of sugar-beet seedlings because they feed on the young roots (Heijbroek, 1971; Winner and Schaüfele, 1967; Dunning, 1972). In England *O. hortensis* Gisin is a pest of crops such as French beans and damage to the seedlings of this crop by *O. fimetarius* (L.) and *O. ambulans* (L.) has been reported (Edwards, 1962); *Onychiurus* damage to sugar beet was first noticed in 1958 and has occurred occasionally since (Jones and Dunning, 1972). This paper records some observations on *Onychiurus* spp. in March and April, 1973, when many were found in the root zone of sugar-beet seedlings in a field at Broom's Barn Experimental Station. Their distribution in and between the crop rows is given for other field trials in 1971-74 and observations on O. armatus feeding on seedlings in the laboratory are reported.

MATERIALS AND METHODS

Sugar-beet seed (cv. Amono), without insecticide treatment, was sown 5 cm deep at 3.8, 11.4 and 22.9 cm spacings in rows 50 cm apart on experimental plots of 0.01 ha in four randomized blocks at Broom's Barn on 15 March, 1973. Many O. armatus were present in a pre-sowing soil sample from the trial area. To assess their distribution in relation to the sugar-beet seed, and to study any root damage, soil samples were taken on two occasions.

On 26 March, before the seedling cotyledons had emerged, 12 soil cores (5.9 cm diameter x 10 cm depth) were taken in the rows where the seed had been sown and another 12 cores midway between the rows in random locations over the whole trial. The Onychiuridae, principally O. armatus and some O. fimetarius, were extracted by crumbling the soil above a Ladell vessel containing water and a layer of petroleum spirit; the liquids were then poured through a fine sieve and the animals, which adhered to the surface of the petroleum spirit, were picked off with a fine brush and numbers per soil core recorded. On 17 April, when approximately 30 - 40 per cent of the seedlings showed above the soil surface, 24 in-row samples of five cores (2.5 cm diameter x 10 cm depth) were taken, each core was taken centred over and including one seedling, together with 24 similar-sized samples midway between the rows (between-row samples). The soil cores were located at random within each plot and the animals extracted by the method previously described. Seedlings included within the in-row samples were removed during the extraction procedure and the roots later inspected for damage.

To ascertain that onychiurid Collembola were the primary cause of the type of damage with which they were associated in the field, some observations were made in the laboratory. One hundred and eighty O. armatus, collected from a field where they were suspected of damaging sugar-beet seedlings, were introduced into a polythene box (17.8 x 12.4 x 4.6 cm) containing a 3-cm layer of moistened, oven-sterilized soil. Ten 'raw' and 10 pelleted sugar-beet seeds (cv. Amono), pre-germinated until radicles were visible, were 'sown' in two parallel rows in small depressions in the soil surface. The polythene lid was snap-fitted and the box kept in darkness in a constant-temperature room at 7°C. Collembola on the seedling roots were counted six times at regular intervals over a period of about eight days until the radicles had grown to 2-3 cm long. The seedlings were removed, washed on a sieve and scored for root damage on a 0-5 scale (0 - no damage, 5 = severe damage and recovery was unlikely). Eight successive sowings were included in the experiment; at each, fresh pre-germinated seeds were introduced and observations repeated as for the first. At the end of the experiment the number of O. armatus left in the box was counted.

R E S U L T S

The numbers of Onychiurus spp. per soil core cannot be directly compared between field samples taken on 26 March and 17 April because of the difference in the size of the sample units; the former consisted of single large cores whereas the latter was of several smaller cores bulked together.

The in-row soil samples taken on 26 March (Table 1) had significantly more <u>Onychiurus</u> spp. than the between-row samples ($P < 1\%$). The in-row sample cores included, by chance, some with a germinated seed and some without; this probably contributed to the wide range of from 9 to 98 <u>Onychiurus</u> spp. per core and the aggregation around individual germinated seeds was therefore underestimated. In the soil samples taken on 17 April numbers of <u>Onychiurus</u> spp. in the row and between the rows did not differ significantly, nor did numbers around seedlings.

TABLE 1

<u>Comparison of numbers of Onychiurus spp. in soil samples on two dates in 1973.</u>

Sampling position	Mean number per sample unit	
Date collected	26 Mar.$^{+}$	17 Apr.$^{++}$
In row	37.1	15.8
Between row	11.7	11.8
't'	3.1	1.4
P	**	n.s.

$^{+}$ sample unit composed of one core (5.9 x 10 cm)

$^{++}$ sample unit composed of five cores (2.5 x 10 cm) bulked

** sample means significantly different at $P < 1\%$

On 29 March, only three days after the soil was first sampled, 40 germinated seeds collected from the experimental plots had radicles

0.3 - 1.3 cm long; two were severely damaged, with many <u>Onychiurus</u> spp. in the vicinity of the damaged radicles. Plate I shows a seedling with small, round, sharply defined pits on the hypocotyl and complete destruction of the radicle and root cap. On 17 April primary roots on the surviving seedlings were up to 12 cm long; a few had pits in the root cortex and excised lateral roots at 5 - 10 cm below the soil surface, and the most typical one is shown in Plate II. Plate III shows five seedlings damaged by <u>O. armatus</u> in the laboratory and Plate IV the effect of damage on seedling growth compared with a control. The distinct holes in radicles and hypocotyls are very similar to those found on seedlings taken from the field; small holes were also sometimes seen on the cotyledons of the laboratory seedlings.

Table 2 records the damage score per seedling for each sowing in the laboratory and is a mean for the 20 seedlings from both 'raw' and pelleted seed. The number of Collembola observed on the seedlings is a mean per seedling from six observations.

TABLE 2

<u>Damage to sugar-beet seedlings in the laboratory</u>

'Sowing'	1	2	3	4	5	6	7	8	Mean
Mean damage score per seedling	1.5	2.4	2.0	2.4	1.2	2.8	1.2	2.8	2.1
Collembola per seedling per observation	0.5	0.9	0.9	0.6	0.5	0.6	0.2	0.9	0.6

The results show that the numbers feeding on the seedlings remained fairly constant throughout the observation period and

PLATE I Severe hypocotyl damage and excision of sugar-beet radicle by *Onychiurus* spp. x 22

PLATE II Damage by *Onychiurus* spp. to primary root and excision of the laterals of a sugar-beet seedling x 3

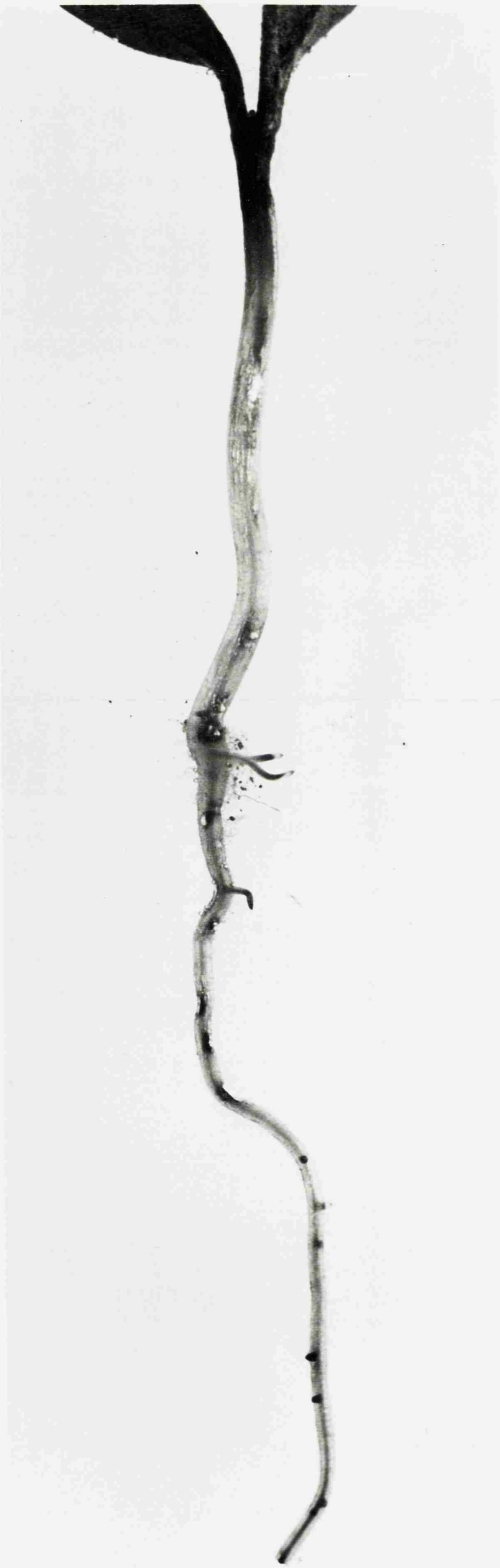
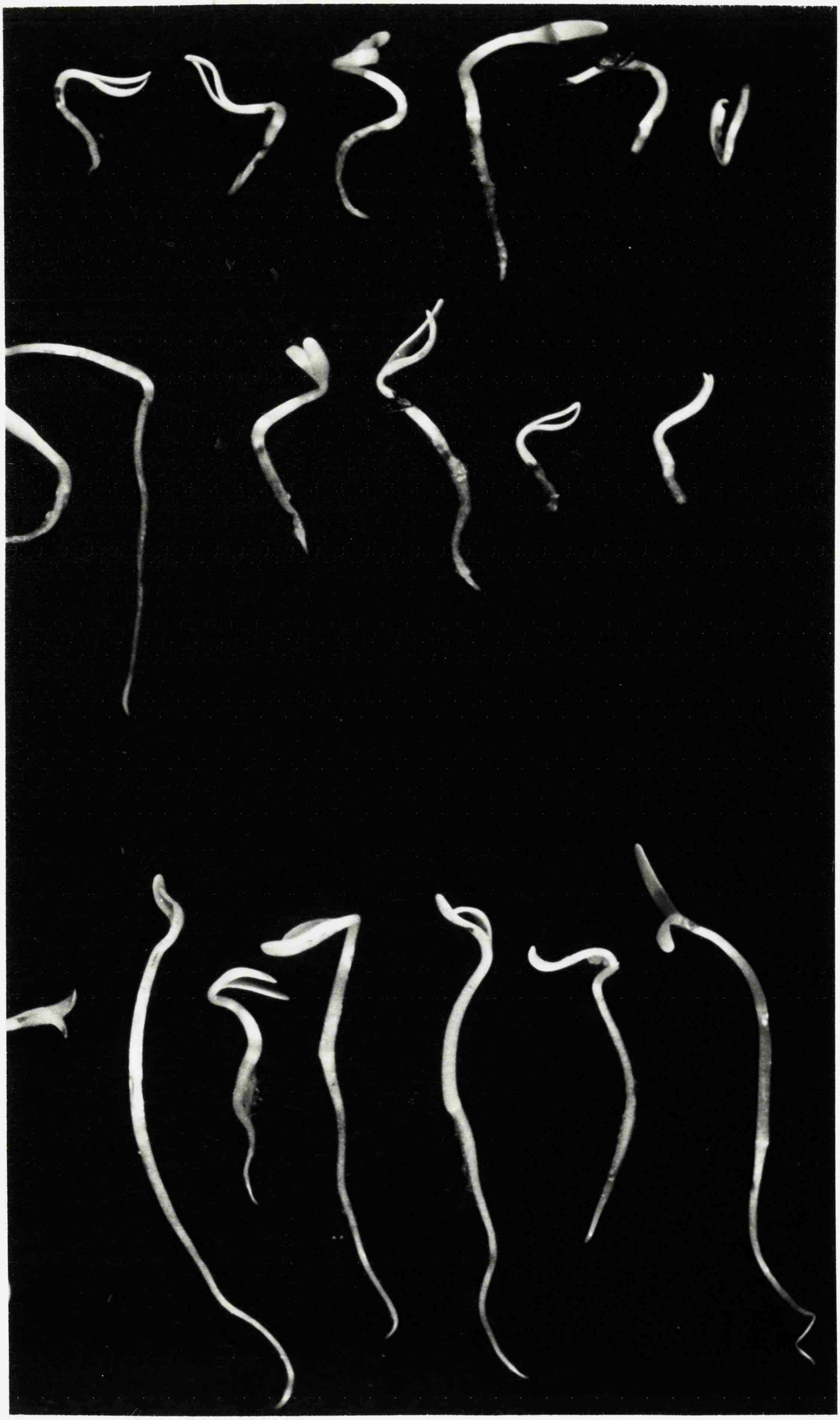


PLATE III Sugar-beet seedlings damaged by *Onychiurus armatus* in the laboratory

PLATE IV The effect of damage by *O.armatus* on the growth of sugar-beet seedlings x 3

Damaged seedlings upper two rows

Undamaged seedlings bottom row



that seedling damage was consistent for every 'sowing'. Of the 180 O.armatus originally introduced in June, 163 were present and alive in the following November.

Soil samples taken at other sites from 1971 to 1974, when the seedlings were in the cotyledon or early rough-leaf stages, indicated that Onychiuridae occurred in significantly greater numbers in the seedling root zone than in the soil between the rows (Table 3), but the numbers in individual seedling root zones were not found to vary significantly with seed spacing (Table 4).

Table 3

The effect of sampling location on the numbers of *Onychiurus* spp. in the soil at three sites in 1972-1973, after emergence of the seedlings.

Sampling position	Mean No. of *Onychiurus* spp. per sample unit Stalham+ Norfolk 28.4.72	Kettering++ Northamptonshire 24.4.73	Littleport++ Cambridgeshire 2.5.73
In row	18.1	30.9	9.4
Between row	6.4	12.3	5.0
S.E.D.	2.9	4.1	1.8
P	**	***	*

***, **, *, sample means significantly different at P <0.1%, 1.0%, 5.0% respectively.

\+ sample unit composed of one core (5.9 x 10 cm)

++ sample unit composed of eight cores (2.5 x 10 cm) bulked

TABLE 4

The effect of different seed spacings on the numbers of onychiurid Collembola around seedlings at five trial sites, 1971-1974.

Seed spacing (cm)	1 Broom's Barn[+] 27.5.71	2 Broom's Barn[+] 11.5.72	3 Kettering[++] 24.4.73	4 Little-port[++] 2.5.73	5 Stalham[++] 6.5.74
	Mean number of Onychiuridae (sites 1 & 2) and *Onychiurus* spp. (sites 3-5) per sample unit.				
3.8	5.1	14.5	35.3	9.5	15.6
11.4 (except site 1=7.6 cm)	5.9	5.5	29.1	7.8	22.3
22.9	6.6	22.3	28.3	11.0	18.9
S.E.D.	1.20	9.0	7.8	3.0	7.6
P	n.s.	n.s.	n.s.	n.s.	n.s.

\+ sample unit composed of one core (5.9 x 10 cm)

++ sample unit composed of eight cores (2.5 x 10 cm) bulked

D I S C U S S I O N

Top-soil samples (10 cm depth) taken at experimental sites throughout East Anglia from 1971 to 1974 indicate that *Onychiurus* spp. are present in most arable soils but numbers vary considerably; for instance, in March and April 1974, populations in seven fields varied from 0 to 5.4 million per acre (0 - 13.3 million/ha). At most sites *Onychiurus* spp. occurred with other pests in the root zone of seedlings; whilst *Onychiurus* spp. are able to cause primary damage, these other accompanying pests such as millipedes, wireworms or pygmy beetles can also do so. Therefore responses to insecticide treatment cannot usually be ascribed to the control of one pest. However, at Broom's Barn in 1973 and at Stalham in 1974, infestations almost exclusively of *Onychiurus* spp. were present and caused damage to the roots before the arrival of other pests.

Populations of soil-inhabiting insects are difficult to estimate owing to their vertical migrations in response to temperature (Dowdy, 1944) and other changes. Blaniulid millipedes have been found to move up into the seedbed in sugar-beet fields in spring (Pierrard, et al, 1963). The movement of Onychiuridae in the soil was recorded by Edwards and Lofty (1971), who found peak numbers in the top 15 cm of soil during April. Heijbroek (1971) recorded that Onychiurus spp. hibernated below the plough layer, down to 70 cm depth, and migrated into the seedbed in March-April when the soil temperature rose to 5°C; despite the low temperature they were very active until the beginning of May, when the drying of the soil drove them deeper. At Broom's Barn in 1973, Onychiurus spp. damaged the roots of sugar beet, especially the seed radicles, in late March when the top 10 cm of the soil was moist and at a temperature of 3-11°C. As the seedlings grew, damage to tissue deeper in the soil, especially the small lateral roots, was probably as much a consequence of the distribution of susceptible tissue suitable for food as it was of the Onychiurus spp. being driven deeper by the increasing temperature and/or dryness of the top soil.

Onychiurus spp. has been shown to aggregate in the row as with millipedes, long-established pests of the crop, but whereas numbers of millipedes per seedling root zone have increased with spacing (Baker, 1973), numbers of Onychiurus spp. have not. However, examination of seedlings roots at the Stalham site in 1974, where soil samples were taken, showed that the amount of damage can be significantly less on the narrowest than on the widest spacing as sometimes recorded when millipedes have been the cause (Baker, 1975). These findings imply that seed spacing was, at some time, affecting numbers of Onychiurus spp. around the seedlings which was not detected by sampling and thus its timing may be important, especially if one considers that spacing effects on soil-inhabiting pests would be less difficult to detect at a time when aggregation in the row was greatest. The time at which this occurs may vary with the different pests; with millipedes it coincides with a peak in numbers when the seedlings are advanced (Baker, 1974) but with Onychiurus sampling has shown that it may be before they have emerged.

The similarity of Onychiurus behaviour to that of other soil-inhabiting pests also extends to their aggregated distribution pattern (Glasgow, 1939). Soil sampling has shown that, as for millipedes, the degree of aggregation increases with the density of individuals (Baker, 1975). Therefore, in a crop suffering from damage, the pattern of damage will reflect their distribution; some seedlings, as laboratory observations confirm, may have large feeding aggregates around each root and suffer severe damage whilst adjacent seedlings may remain undamaged because of few or no Onychiurus.

Commercial sugar-beet seed supplied to growers during the years considered was treated with 0.2 per cent dieldrin (w/w), except that 5 per cent was treated with the same amount of methiocarb in 1974 (Hull, 1974). Untreated seed was sown at our sites where sample location or seed-spacing effects were investigated. In 1972, dieldrin treatment did not prevent young seedlings of a commercial sugar-beet crop at Stalham (Norfolk) from dying, and large numbers of live *Onychiurus* spp. were found around the damaged roots. Field trials in 1974 showed that dieldrin or methiocarb seed treatment can increase seedling establishment slightly and significantly decrease root damage but not significantly affect the numbers of live or dead *Onychiurus* spp. in the root zone (Baker, 1975). Heijbroek (1971) found that seed treatment with lindane or carbaryl can be effective only in cases of light to moderate infestations by *O.armatus*.

Onychiurus spp. may be important pests of sugar beet in England and their true status may easily be hidden by the activities of other pests. They are certainly amongst the most widespread of potential pests that inhabit the soil. Because cases of poor emergence may, in part, be due to unsuspected damage by Onychiuridae there is a need for an accurate survey of populations in different beet-growing areas. Although some work has been done on factors which may stimulate *Onychiurus* populations to increase and on cultural factors which may predispose seedlings to attack (Heijbroek, 1971), further studies are needed, especially in the seed germination and immediate post-germination period.

We thank Mr. B.J.G.Love to Broom's Barn Experimental Station for the photographs and Mr. H. Gough of Jeallot's Hill Research Station for identifying the Collembola.

SOME EFFECTS OF SOIL TYPE AND CROP DENSITY ON THE ACTIVITY AND ABUNDANCE OF THE EPIGEIC FAUNA PARTICULARLY *CARABIDAE*, IN SUGAR-BEET FIELDS

By A.N. BAKER and R.A. DUNNING

Broom's Barn Experimental Station, Higham, Bury St. Edmunds, Suffolk

INTRODUCTION

Sugar beet is grown on soil types covering a wide range of organic content and particle size giving soils of diverse texture and structure (Anon., 1970). Some pest problems are specific to certain soil types whereas others occur over the whole spectrum. For instance, the sand weevil, *Philopedon plagiatum* (Schall), is a pest of sugar beet only in the sandy Breckland areas of East Anglia, damage by pygmy beetle, *Atomaria linearis* Steph., is largely confined to the Fenland regions (Dunning, 1973), whereas the beet flea beetle, *Chaetocnemma concinna* (Marsh), damages sugar beet on the eastern side of the country on a range of soils.

Pitfall traps in sugar-beet fields of two soil types on Broom's Barn farm in 1969 caught considerably different numbers of *Atomaria linearis* and carabids (Chalk and Dunning, 1970). To determine the general epigeic arthropod fauna in sugar-beet crops during the growing season, especially in May, June and July, we trapped on five sites, each with a distinct soil type within a 20 mile (32 Km) radius of Broom's Barn in 1970 and 1971; the catches of both predatory insects and pests of sugar beet were recorded. In 1971 further studies were made in the sugar-beet crop at Broom's Barn to determine whether crop density affected the catch.

METHODS

Pitfall traps

Each trap consisted of an aluminium can* of 450 ml capacity, 63 x 160 mm long with a hole 50 mm in diameter in the screw-cap; the remainder of the cap formed a 7.5 mm lip which was painted matt black. Each can was supported vertically in a 170 mm length of 65 mm internal diameter plastic rainwater pipe sunk in the soil, and supported by the screw-cap so that the lip was level with the surrounding soil. The can contained 40 mm depth of water plus a few drops of wetting agent; arthropods caught drowned quickly and there was no predation by birds.

Traps were placed in the sugar-beet rows soon after sowing, and not later than the emergence of the first seedlings. Later, plants up to 150 mm from the trap were removed to leave a space on either side to allow for expansion of the adjacent sugar-beet tap roots. The traps in the rows were undisturbed by inter-row cultivations, which was done at least two or three times in May-June, nor by hand hoeing of the crop. Traps were changed at weekly intervals with minimum soil disturbance, by sliding the cans from within the plastic tube and resetting fresh ones.

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*Metal Box Company Limited, Oddicroft Lane, Sutton-in-Ashfield, Notts. NG17 5FS

Sites and location of pitfall traps

Soil type and epigeic soil fauna

In 1970 and 1971 pitfall traps were placed in a field on one of each of the following contrasting soils - loamy coarse sand, very fine sandy loam, calcareous silty loam, peaty loam or loamy peat, and clay loam (Table 1). The fields chosen were matched for soil type and, where possible, similar crop rotations so as to be comparable for both years. In 1970 four pitfall traps were placed in each field but six per field in 1971. Half of the traps were located approximately 18 m from the field boundary (hedge, shelter belt of trees, or ditch) and half near the centre of the field, 49-109 m from the edge, depending on field size. In 1970 each trap pair was arranged on a line at right angles to the field boundary and 11 m apart, whereas in 1971 the traps were arranged in a series of three set 11 m apart parallel to the field border and near the centre of the field.

(Table 1 here)

Crop density studies

In 1971 two pitfall traps were placed 5.5 m apart, centrally in plots of 10 x 15 m (0.16 ha) in a 4 x 4 Latin Square experiment at Broom's Barn (Dunholme Field) to test the effect of different sugar-beet plant densities on the catch of carabid beetles and other epigeic soil fauna. Treatments were:

1) Plots with bare soil (not sown and kept free of weeds)
2) A sparse plant population (25,000 per ha), surplus young plants being hoed out on 3 June.
3) A normal plant population (75,000 per ha)
4) An insecticide treatment (results not reported in this paper).

TABLE 1. DETAILS OF PITFALL TRAPPING SITES, 1970 AND 1971

Year		Site	Soil series and type	Soil reference name (used in text)	Previous crop	Total no. of traps	Trapping period
1970	A	Barton Mills, W. Suffolk	Worlington loamy coarse sand	sand	Lucerne	4	29.4 - 28.7
	B	Bottisham, Cambs.	Wantage calcareous silty loam	chalk	Barley	4	29.4 - 13.10
	C	Welney, Norfolk	Wisbech complex very fine sandy loam	silt	Wheat	4	29.4 - 3.11
	D	Ely, Cambs.	Adventurer's deep phase, loamy peat	peat	Wheat	4	29.4 - 4.8
	E	Wickhambrook, W. Suffolk	Hanslope clay loam	clay	Wheat	4	6.5 - 28.7
1971	F	Tuddenham, W. Suffolk	Worlington loamy coarse sand	sand	Winter Barley	6	26.4 - 2.8
	G	Burwell, Cambs.	Wantage calcareous silty loam	chalk	Wheat	6	26.4 - 4.10
	H	Welney, Norfolk	Wisbech complex, very fine sandy loam	silt	Barley	6	26.4 - 4.10
	I	Little Downham, Norfolk	Adventurer's deep phase, peaty loam, sandy	peat	Onions	6	26.4 - 9.8
	J	Ely, Cambs.	Hanslope very fine sandy clay loam	clay	Wheat	6	26.4 - 2.7
	K	Broom's Barn, W. Suffolk	Fine sandy loam		Wheat	32	2.6 - 11.8

RESULTS

Soil type effects

In East Anglia, on the five sites each year with contrasting soil, the most common carabid species trapped were Bembidion lampros, Feronia melanaria, Harpalus rufipes and Amara apricaria during the trapping periods 29 April to 28 July 1970, and 26 April to 26 July 1971 (Table 2). These four species formed 53% and 16%; 40% and 21%; 2% and 19%; and 1% and 23% respectively of the total catch of Carabidae in the two years; the seasonal occurrence of the first three on the chalk and silt sites is shown in Fig. 1. Bembidion lampros was the dominant carabid in May but Feronia melanaria and Harpalus rufipes in July and August; Trechus quadristriatus numbers peaked in September at the three sites where trapping was extended until October (Fig. 1).

(Table 2 and Fig. 1 here)

Bembidion spp. were caught at all sites but fewest on the sand and chalk; Feronia spp. were common at all except the sand sites, on which none were caught in 1970 and comprised less than 2% of the carabid catch in 1971 (Fig. 2). In contrast, most of the Carabidae caught on the sand sites in both years were Amara apricaria and comprised 80% of the total catch in the trapping period 26 April to 4 August, 1971; it occurred only infrequently on all other soil types despite its observed readiness to fly. In both years traps on the chalk sites caught few Carabidae. Most Feronia spp. were caught on the silt and clay sites. Although the numbers of each species caught usually varied considerably from year to year on the same soil type, the total Carabidae caught over the thirteen week period was similar at 780 and 837 per trap for 1970 and 1971 respectively.

(Fig. 2 here)

TABLE 2. CARABID BEETLES TRAPPED ON DIFFERENT SOIL TYPES

(MAY - AUGUST 1970 and 1971)

Genus Species	% of total catch (1970 + 1971)
Bembidion lampros (Herbst)	29.9
Feronia melanaria (III.)	28.4
Amara apricaria (Paykull)	14.5
Harpalus rufipes (Deg.)	12.4
Agonum dorsale (Pont.)	2.9
Clivina fossor (L.)	2.7
Bembidion obtusum (Serv.)	2.4
Trechus quadristriatus (Schrank)	2.1
Metabletus obscuroguttatus (Duft.)	0.8
Harpalus aeneus (F.)	0.6
Asaphidon pallipes (L.)	0.3
Bembidion ustulatum (L.)	0.3
Feronia madida (Fab.)	0.2
Stomis pumpicatus (Pz.)	0.2
Nebria livida (L.)	0.2
Amara consularis (Duft.)	0.1
Notiophilus biguttatus (Fab.)	0.1
Agonum marginatum (L.)	<0.1
Bembidion femoratum (Sturm.)	<0.1
Calathus fuscipes (Goeze)	<0.1
Calathus melanocephalus (L.)	<0.1
Nebria brevicollis (F.)	<0.1
Synuchus nivalis (Pz.)	<0.1

FIG.1. SEASONAL OCCURRENCE OF FOUR COMMON *CARABIDAE* ON CHALK AND SILT SITES, MAY - OCTOBER (A mean for 1970 & 1971)

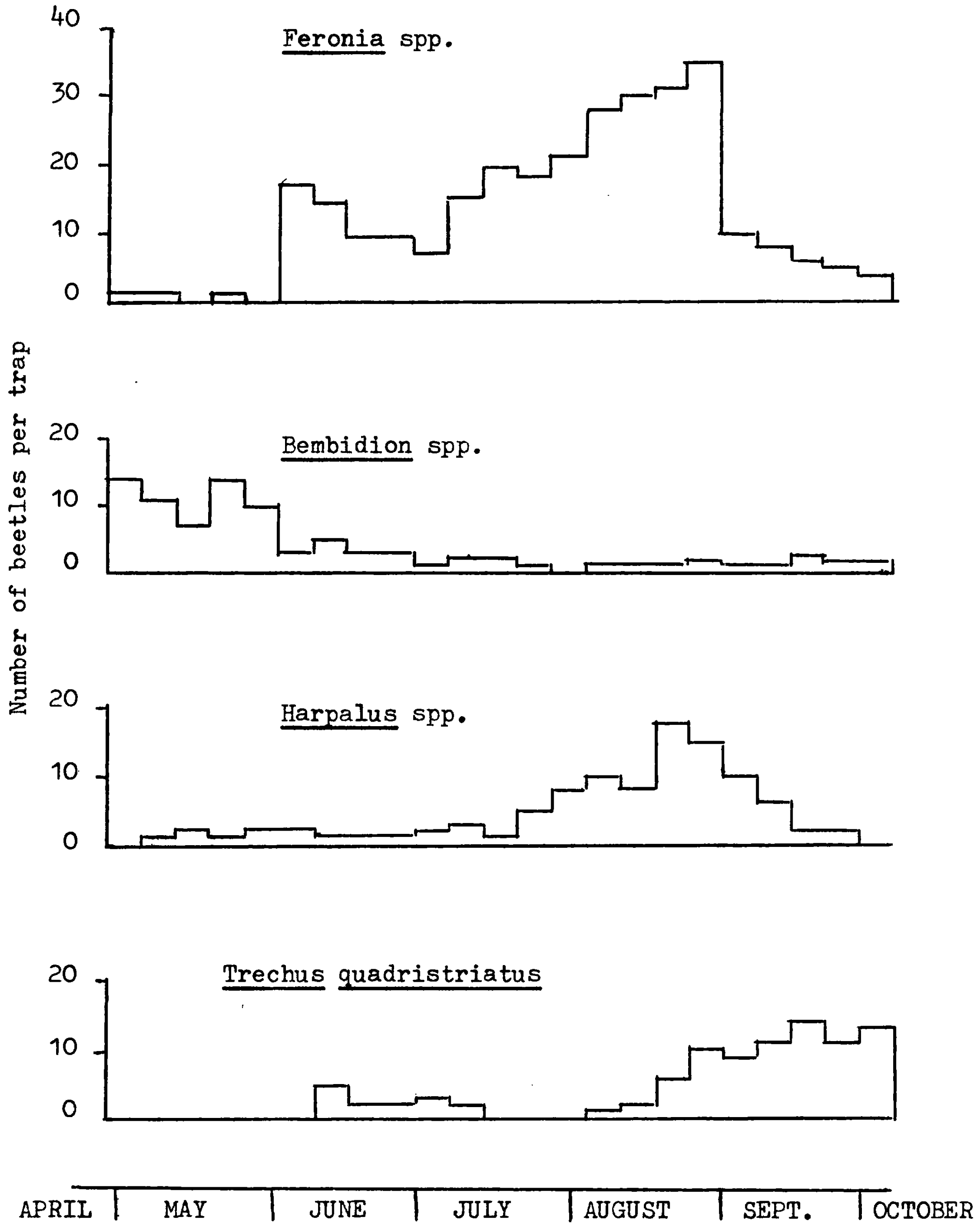
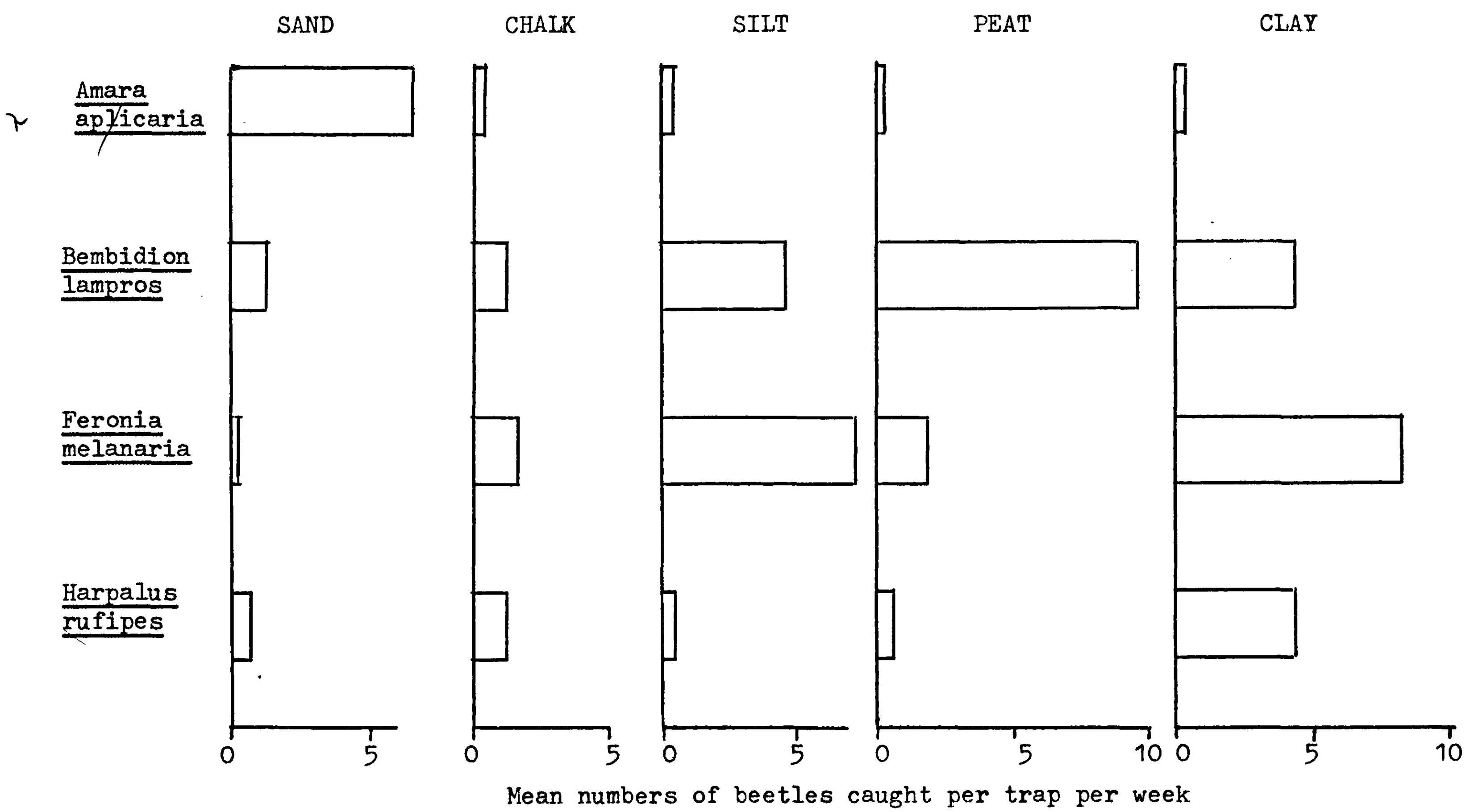


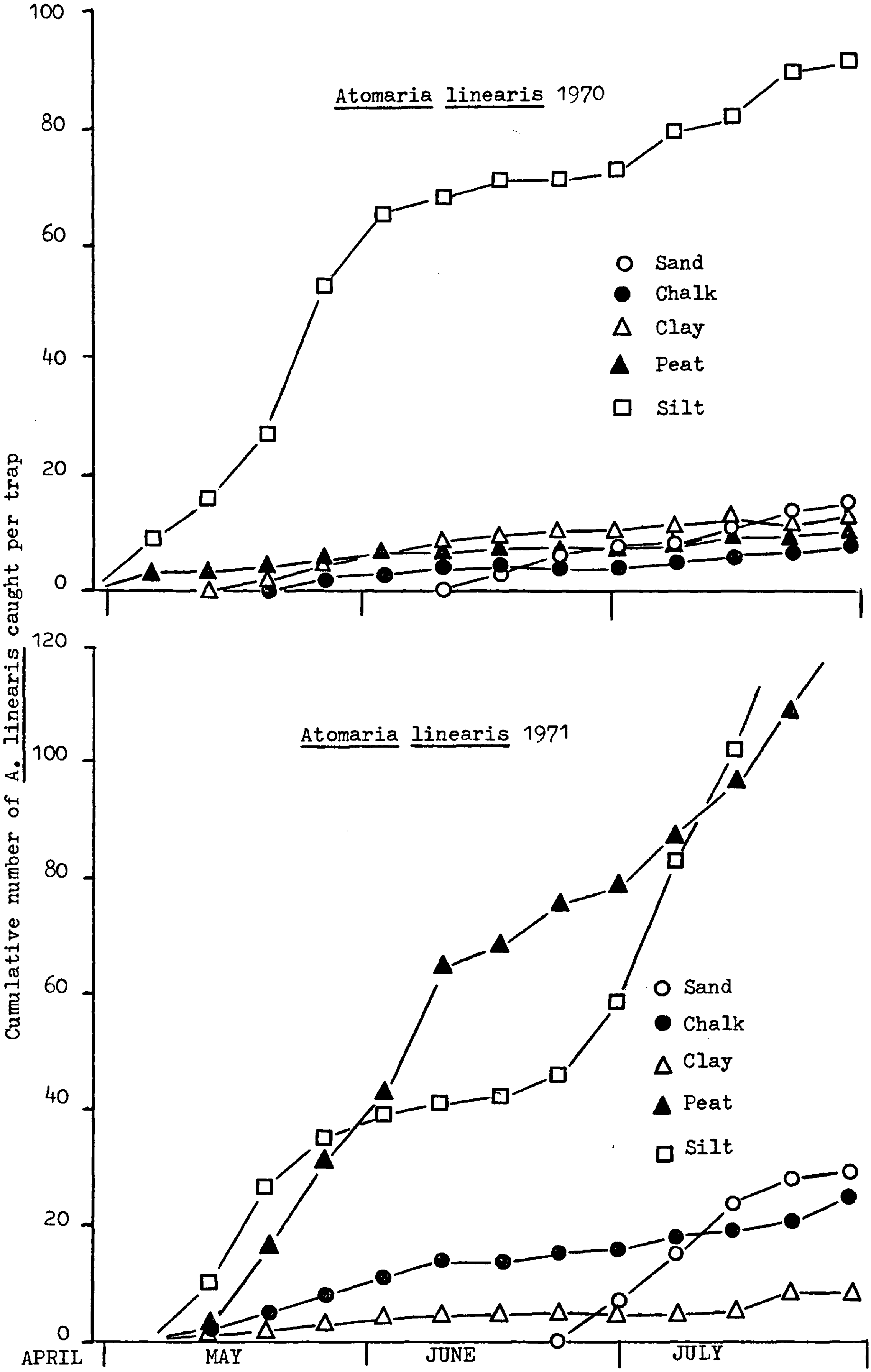
FIG.2. THE DISTRIBUTION OF SOME CARABID BEETLES ON DIFFERENT SOIL TYPES



Other predators were rarely caught; staphylinids and araneids were recorded but not identified. Mean numbers per trap per week on all soil types for 1970 and 1971 were, respectively, 3.2 and 4.2 staphylinids and 2.0 and 1.9 araneids compared with 65 and 70 carabids in the same 12 week trapping period. For each soil type the mean numbers per trap per week of staphylinids and araneids respectively were sand: 2.1, 2.6; chalk: 4.3, 1.3; silt: 4.2, 1.3; peat: 5.0, 1.9; clay: 2.9, 2.6.

Of the sugar-beet seedling pests, *Atomaria linearis* was caught on all soils and more were caught in 1971 than 1970. It was most frequent on the silt and peat; fewer were caught on the sand, chalk and clay sites (Fig. 3). Millipedes were trapped in small numbers during spring on all soil; Polydesmidae, *Brachydesmus superus* (Latzel), *Polydesmus coreaceus* (Porat), and *Polydesmus angustus* (Latzel) were most frequent on the peaty loam, peat, clay and silt soils and the Blaniulid, *Blaniulus guttulatus* (Bosc) was trapped occasionally on the chalk, peat and sand sites. The Iulid, *Brachyiulus pusillus* (Leach) only at the sand sites. Sand weevil, *Philopedon plagiatus*, were trapped in May on the sand sites; to prevent damage the grower sprayed DDT, but later experiments showed little effect from such treatment on the numbers of Carabidae caught. Also peculiar to the sand site in 1970 was an infestation of *Phyllopertha horticola* (L.). Flea beetles (*Chaetocnemma concinna* (Marsh) and *Phyllotetra* spp. were caught at all sites in both years but in very small numbers, never exceeding two per trap per week for the three months May to July, and showed no consistent relation to soil type.

FIG.3 CATCH OF ATOMARIA LINEARIS 1970 AND 1971 ON DIFFERENT SOIL TYPES



TRAP LOCATION EFFECTS

Averaged over all soil types, the pitfall traps placed near the field centre caught 12% more carabids in 1970 and 19% more in 1971 than the traps near the field boundary; these latter traps were at least 14 m from the boundary and crop growth was identical to that at the field centre. For both years the largest differences were at the chalk and silt sites, respectively 22% and 27% more caught centrally than near the boundary; Bembidion lampros, Feronia melanaria, and Harpalus rufipes were the dominant species on these soils. In 1971, when the catch difference was greatest, there were 37% more Amara apricaria, 6% more Harpalus rufipes, 13% more Feronia melanaria but 8% fewer Bembidion lampros in field centre traps averaged for all soil types. For each site, differences in catch for each of these species were non-significant, using the Mann-Whitney U test to compare medians, except that at the sand site significantly more Bembidion lampros ($U = 37.5$, $P < 5\%$) were caught in traps sited near the field boundary.

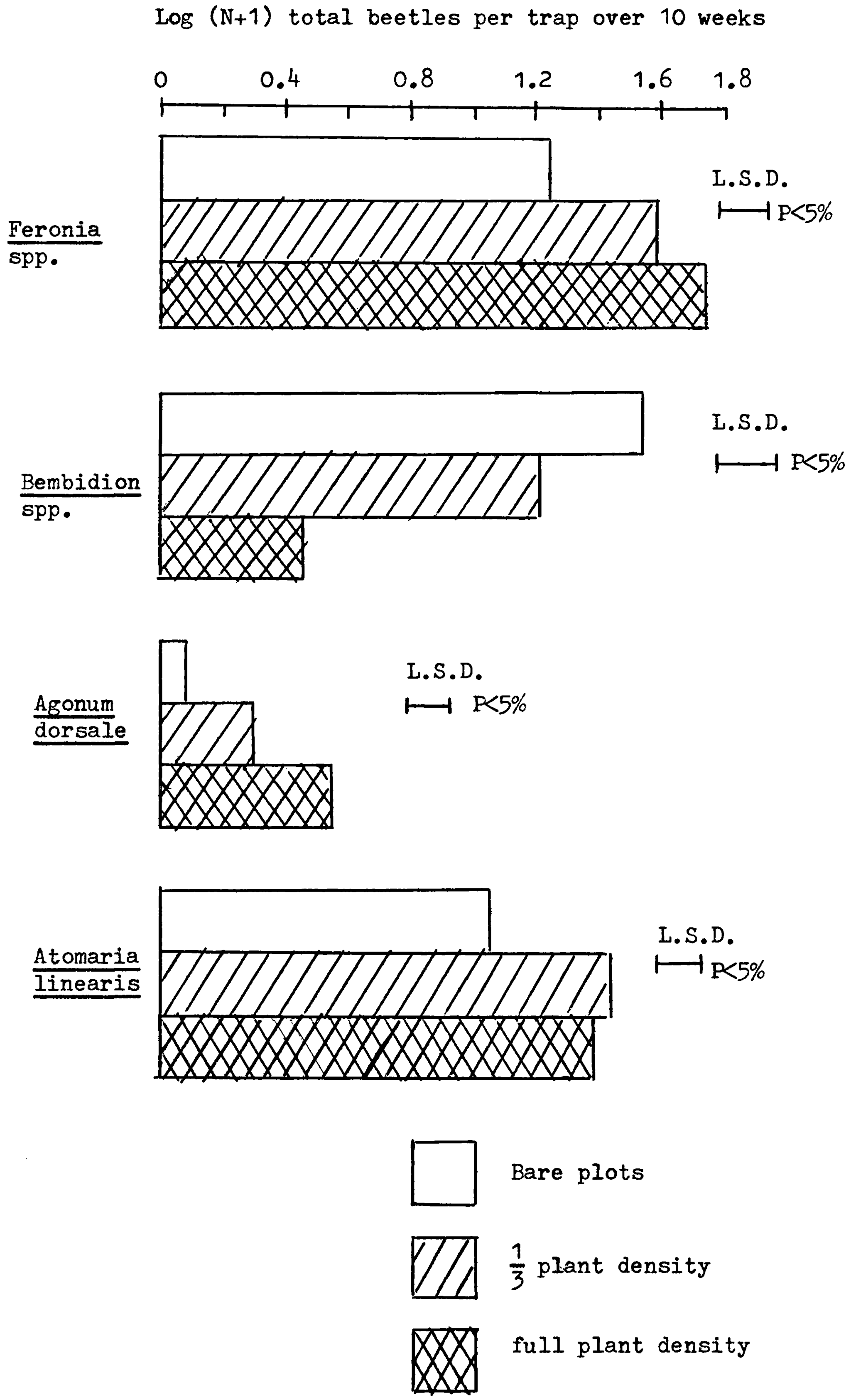
In contrast to most carabids, most Atomaria linearis were usually caught in traps nearest the field boundary, on average of all soils 50% more in 1970 and 19% more in 1971, probably due to the effect of a windbreak on the immigrating beetles.

CROP DENSITY STUDIES

At the start of the experiment, before hoeing was done to differentiate bare plots and those with low plant population, it was confirmed that the carabids were uniformly distributed over the whole experimental area; trap catches did not differ significantly. Treatment replication was an additional safeguard against subsequent heterogenity and the experiment was sited towards the middle of a 17 ha field to minimise any possible boundary effect. Fig. 4 shows that the catch of carabidae from 2 June until 11 August 1971 varied with sugar-beet plant population. Most Feronia spp. and Agonum dorsale were caught on plots with the normal plant population and fewest on the bare plots; with Bembidion spp. the converse occurred. With Atomaria linearis, an immigrant species, the catch did not differ between plots with normal and one third plant density, but was significantly less on the bare plots.

(Fig. 4 here)

FIG.4. EFFECT OF SUGAR BEET PLANT DENSITY ON THE PITFALL TRAP CATCH OF SOME SOIL-INHABITING BEETLES. MAY-JULY 1971



DISCUSSION

Briggs (1961) compared the technique of pitfall trapping to soil sampling and concluded that the number of carabids caught in pitfall traps is related more to activity fluctuations than absolute population changes. Greenslade (1964) recognised that susceptibility to being caught may vary with different species of carabid according to size, behaviour and strata in which they are active in the ground vegetation. However, these restrictions do not inhibit the use of limited quantitative comparisons between sugar-beet crops at different sites, sown at the same time and growing at similar rates, such as the first part of the present study describes.

The dominance of _Bembidion_ spp. (mainly _B.lampros_) in pitfall trap catches from arable land in April and May is recorded by Davis (1968) and Critchley (1972). _B.lampros_ emerge from pupae between July and September, are particularly active as adults during winter, and breed in the spring; in contrast, _Trechus quadristriatus_ numbers usually reach a peak in September and October and this species is described as an 'autumn breeder' (Mitchell, 1963). The increasing abundance of _Feronia_ spp. (particularly _F.melanaria_), and of _Harpalus_ spp. (mainly _H.rufipes_) in our June, July and August catch is probably indicative of true population peaks; the main emergence of _Feronia melanaria_ from the pupa occurs in June (Briggs, 1965). However, pitfall trapping describes only periods of greatest numbers, the weekly variation being controlled by temperature (Briggs, 1961).

Although the aim of the present study was to assess the epigeic fauna of sugar-beet fields, results of trapping over a wide range of soil types with similar vegetation showed that carabids are more abundant on clay, silt and peat soils than on 'chalk' or sandy soils. Tischler (1955) attributed some of the differences in carabid fauna on sandy and loam soils to food preferences, making the distinction between those species which are mainly phytophagous and those zoophagous. Many of the carnivorous species such as Bembidion lampros and Trechus quadristriatus feed on small, active arthropods such as collembola found at the soil surface (Mitchell, 1963) and the relative abundance of this food may be important in determining the selection of their habitat. Amara spp. which feed to a large extent on seeds and other vegetable matter (Lindroth, 1974), were the most prevalent carabid on the sand sites, whereas Feronia melanaria, which is mainly carnivorous (Skuhravy, 1958), were notably scarce at these sites but most numerous on silt and clay. In 1973 studies were made on a sand site comparing pesticide and non pesticide treated plots (Baker and Dunning, 1975); as in the 1970 and 1971 observations, F.melanaria was relatively scarce.

The dominance of only a few carabid species in the traps may have been due to their activity but is more likely to have been a reflection of the time of year when adults were numerous. Since soil samples were not taken and larvae were seldom recorded in the traps, the composition of the carabid fauna may be an underestimate. Further, the numbers of the few species which fly, such as Amara apricaria, would be overestimated. Some species, such as Bembidion lampros and Feronia melanaria, were more common in traps in 1970, whereas Harpalus rufipes and Amara apricaria were trapped in greater frequency in 1971. The large variation in their percentage representation between the two consecutive years may not merely be due to the incumbence of trapping at different sites, albeit of the same soil types, but may be related to the different climatic conditions in the respective trapping periods. In the May-June period in 1970 the weather was exceptionally dry and warm (Anon, 1971), which contrasted sharply with the same period in 1971 which was wetter and colder than average (Anon, 1972).

Atomaria linearis numbers caught on the different soil types probably reflects habitat preference because the beetle penetrates deep into the soil when conditions are adverse. The time of arrival of A.linearis in the crop largely determines the extent that their feeding damages the young seedlings; they were first trapped at similar times on the chalk, clay, peat and silt sites but at least six weeks later on the sand sites, when the seedlings were well established. Numbers caught was reflected in the extent of damage to seedling roots, which differed in 1971 at the different sites (Baker, Dunning and Winder, 1972).

In order to help explain variations in the catch from habitats differing in either the extent or type of vegetation (Rivard, 1966), it is useful to recognise the factors which affect the behaviour of the beetles under field conditions. In the present study the catch of different carabids in the traps varied significantly with density of the surrounding crop. The greater frequency of trapping Feronia spp. on the plots with most cover may be related to their nocturnal habit (Chalk and Dunning, 1970) and hence possibly to longer periods of activity in the shaded conditions provided by the sugar-beet foliage; their greater abundance, as determined by soil sampling, in soil covered with dense grass or annual weeds was noted by Briggs (1965). The large catch of Bembidion spp. on the bare plots may be attributed either to their diurnal habit or their greater abundance on bare soil as demonstrated by Critchley (1968) using a capture-mark-recapture technique.

Thompson and Smith (1972) found that the density of mini-cauliflowers affects the number of cabbage root fly eggs laid per unit area. Since some carabids are known to be important predators of cabbage root fly (Coaker and Finch, 1971) and their activity and numbers can be affected by crop density, then their predatory efficiency may also be influenced (Wheatley, pers. comm.). The investigations reported in this paper indicate that if intense adult activity can be equated with predatory behaviour, then the value of certain species may vary according to both season and extent of ground cover; It may well differ also, because of numbers, with soil type.

We have shown that several carabid species consume aphids and climb over sugar beet plants in search of them (Dunning, Baker and Windley, 1975). In these pitfall trapping studies on different soil types both apterous and alate green aphids were trapped in 1970 but numbers were not recorded; in 1971 they were trapped at all sites, but by far the most on the sand, 2.4/trap/week v 0.2 - 0.4 on the other sites (mainly Myzus persicae (Sulzer), Aulacorthum solari (Kltb.) and Macrosiphum solanifolii (Ashm.) but species not recorded. No Aphis fabae Scop. See Heathcote, Dunning and Wolfe, 1965). In the same three month period, May to July, coccinellids were caught only on the sand site in 1970 (0.2/trap/week) but at all sites in 1971, the most on the sand (1.4/trap/week v 0.1 - 0.2 on the other sites); species were not identified but were mainly *Coccinella septempunctata*. These numbers are remarkably few in comparison with the 65-70 carabids/trap/week caught in the same period. Observations in 1974 suggested that predators other than coccinellids may be implicated in the control of aphids infecting sugar beet (Baker and Dunning, 1975).

Further studies are needed on the importance of carabid beetles in the sugar beet crop, especially in relation to aphid infestation, and the effects on them of commonly applied insecticide treatments.

SUMMARY

1. Pitfall trapping in sugar-beet crops in East Anglia in 1970 and 1971 showed that Carabidae were much more active and abundant than other epigeic predators.

2. *Bembidion lampros*, *Feronia melanaria*, *Amara apricaria* and *Harpalus rufipes* were, on average, the commonest species.

3. On the five soil types studied there were differences in both species composition and numbers of Carabidae, and especially of sugar-beet seedling pests.

4. Trap location within the field had slight, but mostly insignificant effects on numbers of Carabidae trapped.

5. Density of sugar-beet plants affected differentially the numbers of the most common carabid species; Fewest *Feronia* spp., *Agonum dorsale* and the cryptophagid *Atomaria linearis*, but most *Bembidion* spp., were trapped on bare soil.

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Carabids in sugar beet crops and their possible role as aphid predators

By R. A. Dunning, A. N. Baker and R. F. Windley

Broom's Barn Experimental Station, Higham, Bury St Edmunds, Suffolk

Carabid beetles have been studied by many workers interested in the predators of agricultural pests. The value of the genera *Bembidion* and *Trechus* for the control of cabbage root fly, first recognized as important predators by Wright (1956), was demonstrated by Wright, Hughes & Worrall (1960). Other pests have been shown to be part of the diet of carabids on arable soils, e.g. wireworms (Fox & Maclellan, 1956), slugs (Poulin & O'Neill, 1969), and egg pupal stages of wheat bulb fly (Ryan, 1973*a*, *b*). Insecticides applied to soil or foliage to control pests in the field can have adverse effects on their predators. Dempster (1967) reported increased infestation of *Pieris rapae* caterpillars following DDT application and attributed this partly to the effects of the chemical on both the feeding behaviour and population size of carabids on the sprayed plots. Some observations on the sugar-beet crop have also suggested that insecticides may be affecting the predatory fauna. A significant 16 % increase in virus yellows infection of a sugar-beet seed crop followed a DDT spray applied to control aphids in the previous autumn (Hull & Gates, 1953). Similarly, increased virus yellows in the sugar-beet root crop followed the spraying of trichlorphon to control *Pegomya betae* Curt. (Dunning & Winder, 1965) and mevinphos to control aphids (unpublished data).

The surface-inhabiting invertebrate fauna of sugar beet fields was studied by pitfalls trapping on two soil types at Broom's Barn in 1969 (Chalk & Dunning, 1970) and on five different soil types at outside centres in both 1970 and 1971. Pitfall traps, consisting of screw top aluminium cans 160 × 63 mm with a 50 mm diameter aperture in the top and containing a small quantity of water, were placed in the sugar beet rows shortly after sowing at the end of April; trapping continued until at least the end of July (Dunning & Baker, 1971; Baker, Dunning & Winder, 1972). The first occcinellids were trapped in early July in 1970, but in early May 1971, numbers averaged less than 0·1 and 0·4 per trap per week for each year respectively. The most common carabid, *Bembidion lampros* (Herbst), averaged 6·3 in 1970 and 2·1 in 1971 per trap per week for the same period and other species were nearly as numerous. Aphididae, trapped but not recorded in 1970, averaged 0·7 per trap per week for 1961 and, like the Coccinellidae, were most numerous on the loamy sand sites where fewest carabids were caught in all three years.

Virus yellows infection is brought into the sugar beet crop by winged viruliferous aphids in May and June; they feed restlessly, perhaps moving to other nearby plants, and produce young. Further spread of infection within the crop from these initial foci is due both to aphids walking on the soil surface from plant to plant or from leaf to leaf, or by short flights of winged aphids (Broadbent, 1952). At this time, in May and June, *Bembidion* spp. are the most numerous carabids in sugar-beet fields, more than ten per trap per week often being caught at some sites. Crop infestation by *Myzus persicae* Sulz. usually reaches a peak at about the end of June, but by *Aphis fabae* Scop. not until mid-July (Jones & Dunning, 1972). *Feronia melanaria* (Illiger) increases in abundance and activity during late June and in July; catches of more than fifty per trap per week were common at the end of July at some sites. The rapid decline in aphid numbers in late July thus coincides approximately with a peak in carabid numbers and activity; the decline is usually attributed to the more obvious and specific predators (such as syrphid larvae, ladybirds and lacewings and their larvae, anthocorids and their nymphs), to parasites, to aphid diseases and to emigration. However, possibly, carabids may contribute to the natural control of aphids on the crop foliage or when wandering on the soil surface, especially early in the season.

Laboratory tests showed that *F. melanaria*, *Bembidion lampros*, *B. femoratum* (Sturm.), *B. quadrimaculatum* (L.), *B. ustulatum* (L.), *Harpalus rufipes* (Deg.), *H. aeneus* (F.) and *Trechus quadristriatus* (Schrank), the only species tested, fed readily on nymphs of *A. fabae* when confined together in Petri dishes; for example, *F. melanaria* consumed 7·8 *A. fabae* per 24 h, in comparison with 6·5 by *Anthocoris* sp. In later tests, colonies of *A. fabae* were established on the leaves and these were consumed in part or whole (Windley, Baker & Dunning, 1973). Similarly, Davis (1973) found that the carabid, *Risophilus atricapillus* (L.) readily feeds on aphids in the laboratory; he also recovered this species from suction samples collected from stinging nettles, demonstrating that it had climbed the plants. Dempster (1967) observed species of *Agonum*, *Amara*, *Bembidion*, *Harpalus*, *Notiophilus* and *Trechus* on brassica plants.

We tested whether several species of carabids could climb sugar beet plants in the laboratory. A complete sugar beet crown foliage was collected from the field and set in a glass chamber. Fluorescent powder (micronized Saturn yellow, Series T) placed around the base of the plant contaminated the feet of beetles put in the chamber; after 18 h in darkness the tracks the beetles left on petioles and laminae could be observed under ultra-violet light. *F. melanaria* was the most active climber, but *Agonum dorsale* (Pont.) and *Amara familaris* were also active, although smaller beetles, such as *B. lampros* and *T. quadristriatus*, were less active, possibly because the powder adhered to their legs and tended to impede their movement. Using a powered suction sampler in the field, *Notiophilus biguttatus* (Fab.), *T. quadristriatus* and *B. lampros* were recovered from sugar beet foliage, but sampling was not extensive and no data was obtained on relative numbers, nor on species present in the field but not on the foliage.

In 1973, a field trial at Broom's Barn attempted to determine the possible importance of carabid beetles in the natural control of aphids infesting sugar beet. Treatments were designed to achieve three different levels of carabid beetle population in experimental plots each 50 × 80 ft (15·3 × 24·4 m), arranged in a 3 × 3 Latin Square. One treatment had barriers which allowed ingress of wandering beetles to a central 20 × 17 ft (6·1 × 5·2 in) trapping and observation area but prevented egress, a method similar to that employed by Wright *et al.* (1960) and later by Coaker (1965). The second treatment, spraying 8 lb parathion/acre (9 kg/ha) and working it into the top 6 in (15 cm) of soil before sowing, killed most existing beetles and larvae in the soil (Edwards, 1972); later entry of non-flying beetles into the sprayed area was considerably decreased by the use of an ingress barrier. The third treatment was a control, with no insecticide or barriers.

Four pitfall traps were set centrally in each plot and left in the field for either 24 or 48 h periods once in each week from May until August. Beetles caught were identified in the laboratory, and live ones returned to the plots on which they were caught.

Consistently fewer carabids were caught on the parathion-sprayed plots than on the controls, significant in 7 out of 12 wk. Conversely, and except for 1 wk, catches were always greater on the plots with egress barriers than on the controls, but significantly so only in 1 wk. Fig. 1 shows the cumulative carabid catch for each treatment from May until August and illustrates the effectiveness of the treatments in establishing different carabid populations. *B. lampros* and *F. melanaria* were the most abundant species over the 12-wk trapping period, averaging 2·4 and 4·1 respectively in four traps per plot during each period. Significantly more of these two species ($P < 0{\cdot}05\,\%$) were caught on the egress-barriered and control plots than on the insecticide-treated plots. The effectiveness of the egress barriers in increasing the density of the beetles in the trapping area over that on the control plots was not as great as expected and only *B. quadrimaculatum* was trapped in significantly greater numbers ($P < 0{\cdot}05$) – in the total of the twelve trapping periods).

Aphids on the sugar beet foliage were counted at six intervals from 11 June to 20 July inclusive, by which time numbers were declining rapidly and a later count was not possible. Inevitably, because of differential colonization, numbers of aphids per plant varied greatly and only very large differences in numbers between treatments were significant. The numbers of apterous *A. fabae* increased rapidly until the 12 July, when a mean number of 200 per plant was found on parathion-treated plots (i.e. those with fewest carabids). Greater numbers of *A. fabae* were found on these plots compared to the control in five of the six counts but differences were significant in only one count. Plots with egress barriers to increase the carabid population, on average, had fewer black aphids than the control in each of the three July counts, but the differences were not significant. *M. persicae* were not so numerous, reaching a peak of only 2·4 per plant in late June, and numbers did not differ significantly between treatments at any of the counts. Plants infected with virus yellows were counted on 17 July and 1 and 14 August. The control plots had 8, 21 and 33 % of infected plants on the three respective dates; on the parathion-sprayed plots there were slightly less (5, 20 and 30) and only half (3, 11 and 13) on the plots with egress barriers and most carabids, but the differences were insignificant.

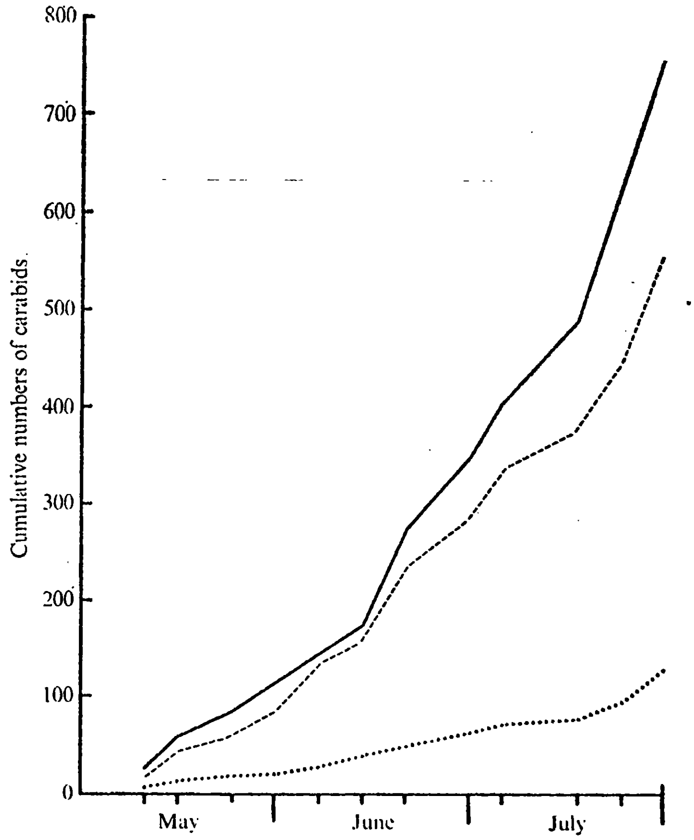

Fig. 1. Numbers of carabid beetles trapped on each of three plot treatments. ——, Egress barrier; ---, control; . . . . ., parathion spray + ingress barrier.

Unfortunately, later virus counts could not be made because the effects of drought masked the virus symptoms.

The experiment did not demonstrate any verified true effect of the different carabid numbers on aphid populations but the results suggest that aphid numbers and virus incidence were being influenced by carabids. It appeared that the effect of carabids on aphid numbers was greater in July than in June but this may be merely the result of aphid multiplication from real but small differences caused earlier.

A large proportion of the sugar beet crop is treated annually with aphicides at least once in the period late May to mid-June and a small but increasing proportion with gamma-BHC and other materials even earlier in the seasons to control seedling pests. It is important to know whether carabids contribute to the natural control of sugar beet aphids, and of other pests; further work is needed, especially on different soil types.

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